L-ARGININE SUPPLEMENTATION IN IUGR AND IT'S EFFECT ON FETAL OUTCOME - A RANDOMISED CONTROL TRIAL

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BONAFIDE CERTIFICATE

This is to certify that the dissertation entitled **"L-ARGININE SUPPLEMENTATION IN IUGR AND IT'S EFFECT ON FETAL OUTCOME"** is the bonafide original work of **Dr. HAJEE ARSHIYA BAREEN** under the guidance of **Dr.K.L.MALARVIZHI, MD.,DGO.,DNB.,** Professor of Department of Obstetrics and Gynaecology, KMCH, Chennai in partial fulfilment of the requirements for MS Obstetrics and Gynaecology branch II examination of the Tamilnadu Dr.MGR Medical university to be held in May 2018. The period of Postgraduate study and training is from June 2015 to May 2018.

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DECLARATION

I solemnly declare that this dissertation **"L-ARGININE SUPPLEMENTATION IN IUGR AND IT'S EFFECT ON FETAL OUTCOME – A RANDOMISED CONTROL TRIAL"** was prepared by me at Government Kilpauk Medical College and Hospital, Chennai, under the guidance and supervision of **Dr. K.L. MALARVIZHI, MD., DGO., DNB.,** Professor, Department of Obstetrics and Gynaecology, Government Kilpauk Medical College and Hospital, Chennai. This dissertation is submitted to **The Tamil Nadu Dr.M.G.R. Medical University, Chennai** in partial fulfillment of the University regulations for the award of the degree of **M.S. (Obstetrics and Gynaecology).**

Place: Chennai

Date: **(Dr. HAJEE ARSHIYA BAREEN)**

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I start this thesis in the name of Almighty God, the most beneficent and forgiving. I thank God for giving me the privilege to learn from the able teachers in my department.

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(Dr. HAJEE ARSHIYA BAREEN)

ABBREVATIONS

AC	-	Abdominal circumference
ACOG	-	American College Of Obstetrics and Gynaecology
AFI	-	Amniotic fluid index
AGA	-	Appropriate for gestational age
AREDV	-	Absent/reversed end diastolic velocities
BMI	-	Body mass index
BPP	-	Biophysical profile
CPR	-	Cerebroplacental ratio
CTG	-	Cardiotocography
DV	-	Ductus venosus
EFW	-	Estimated Fetal weight
eNOS	-	endothelial NO synthase
FDA	-	Food and Drug Administration
FL	-	Femur length
HC	-	Head circumference
HIV	-	Human immunodeficiency virus
IGF	-	Insulin like growth factor
IGFBP	-	Insulin like growth factor binding protein
IUD	-	Intrauterine death
IUGR	-	Intrauterine growth restriction

LBW	-	Low birth weight
MCA	-	Middle cerebral artery
PAPP-A	-	Pregnancy associated plasma protein-A
PI	-	Pulsatility index
PTSGA	-	Preterm small for gestational age
RCOG	-	Royal college of obstetrics and gynaecology
SDVP	-	Single deepest vertical pocket
SFH	-	Symphysiofundal height
SGA	-	Small for gestational age
TORCH	-	Toxoplasmosis, Others(Syphilis, Parvovirus B-19, Varicella Zoster), Rubella, Cytomegalovirus, Herpes
UA	-	Umbilical artery
UV	-	Umbilical vein

CONTENTS

Introduction

INTRODUCTION

Foetal development represents a critical period in humans. The growth of a normal foetus is controlled by a delicate balance of genetic, maternal, placental and foetal factors.

- The genetic drive for the growth
- Environmental factors in uterus
- The supply of growth substrates to the foetus
- Potential of foetus per se to grow

Any alterations in the afore said factors may result in the restriction of growth of the foetus.

Among the intrauterine factors, nutrition plays the most important role in affecting placental and foetal growth. The supply of substrate to the foetus is regulated by maternal - placental factor.

The current view in embryology is that a foetus has an inherent potential to grow into a healthy appropriately sized newborn. However, if there is an imbalance in one or more of these critical growth and development factors, the foetus may fail to achieve appropriate size & weight.

SIGNIFICANCE OF FOCUSSING ON IUGR

Intrauterine growth restriction (IUGR), a condition in which the foetal growth is restricted pathologically in utero, remains a serious health problem; as it affects not only the neonatal period, but also the adult phenotype and quality of life.

IUGR has been defined as the foetal growth rate that is below normal with respect to the growth potential of a specific infant for the respective race and gender of the foetus. It can also be described as a deviation from an expected foetal growth pattern and is usually due to the result of innate reduced growth potential or other multiple adverse effects on the foetus.

IUGR represents the second most common cause of perinatal mortality, after prematurity, and it is related to an increased risk of perinatal complication as hypoxemia, low APGAR scores and cord blood acidemia, with possible negative effects on neonatal outcome. It has been proven by studies that there is an increased risk of premature birth, reduced survival of the neonate and long-term sequelae like impairment of neuro-developmental progress in childhood and insulin-resistance in adulthood, associated with IUGR.

There has been significant association of IUGR with increase in morbidity and mortality in perinatal period and infancy as shown in figure-1. The adverse consequences of growth deprivation in utero extending beyond early years into later life is one of the most worrisome aspects of IUGR.

IUGR AND SGA

A neonate is considered as having normal weight if the birth weight lies between the 10th and 90th percentile with respect to the gestational age, gender and race with no features of malnutrition and growth retardation. The terms “IUGR” and “small for gestational age (SGA)” have been used synonymously, but there exists certain small differences between the two.

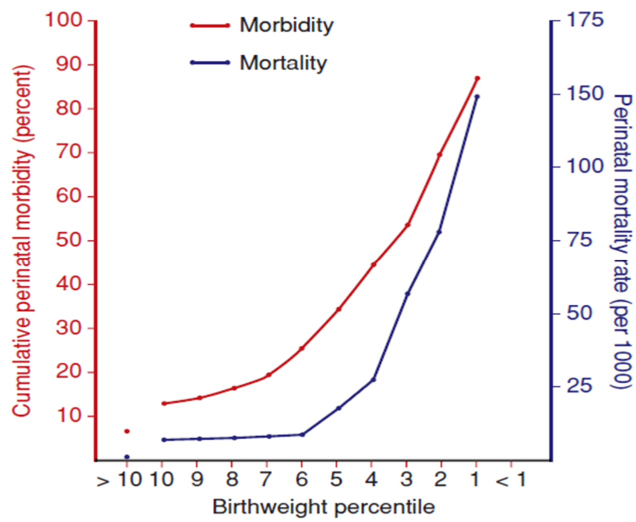


Figure -1 : Relationship between birthweight percentile and perinatal mortality and morbidity rates in 1560 small-for-gestational age foetuses. A progressive increase in both mortality and morbidity rates is observed as birthweight percentile falls. (Data from Manning, 1995.)

The term Small for gestation (SGA) is based on the cross-sectional evaluation of the baby (either prenatally or postnatally), and has been used to denote those neonates whose birth weight is less than the 10th percentile for that particular gestational age or two standard deviations below the norms of the populations on the growth charts. The definition considers only the birth weight without any consideration of the in-utero growth and physical characteristics at birth.

An intra uterine growth restriction (IUGR) is a clinical definition and thereby it applies to those neonates who are born with clinical features suggestive of malnutrition and in-utero growth retardation; with estimated foetal weight falling below the 10^{th} percentile for that particular gestational age of the foetus. However, appropriate for gestational age (AGA) babies can also be labelled as having IUGR if there are features of in-utero growth retardation and malnutrition at the time of birth.

Hence it is important to note that neonates with a birth weight less than the 10^{th} percentile for the gestational age will be small for gestation(SGA), but not an IUGR if they exhibit no features of malnutrition, and a neonate having a birth weight greater than the 10^{th} percentile will be termed an IUGR in spite of being an AGA, if the infants exhibit features of having malnutrition at birth.

Low birth weight (LBW) is a separate entity and it should not be confused with IUGR/SGA, as it is based on the birth weight (less than 2,500 g) of the foetus irrespective of the gestational age, sex, race, and clinical features. However, the terms IUGR and SGA are used interchangeably in medical literature.

A low birth weight baby can be any of the three.

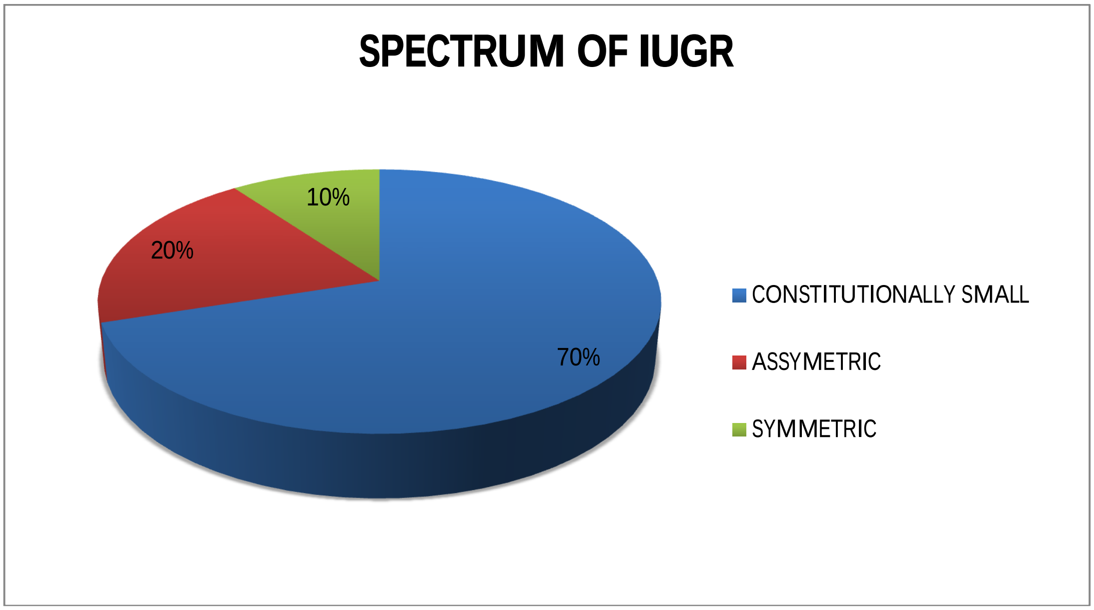


Figure – 2: Spectrum of SGA foetuses

The incidence of IUGR is about six times higher in the developing countries as compared to that of the developed countries. This incidence may further be higher in the lower- and middle-income countries; as many deliveries occur at home with no birth records. The incidence of IUGR is different among various countries, populations, races and it increases with decreasing gestational age.

A large number of IUGR infants, accounting for approximately 75% of all the affected infants, are seen in the Asian continent. This is followed by the African and Latin American continents accounting for 20% and 5% of cases respectively. Country specific rates of IUGR-LBW can be categorized as percentages of all births, as follows: low (<5%), moderate (5-10%), high (10-15%) and very high (>15%). India stands second highest in the incidence of IUGR among the South Asian countries.

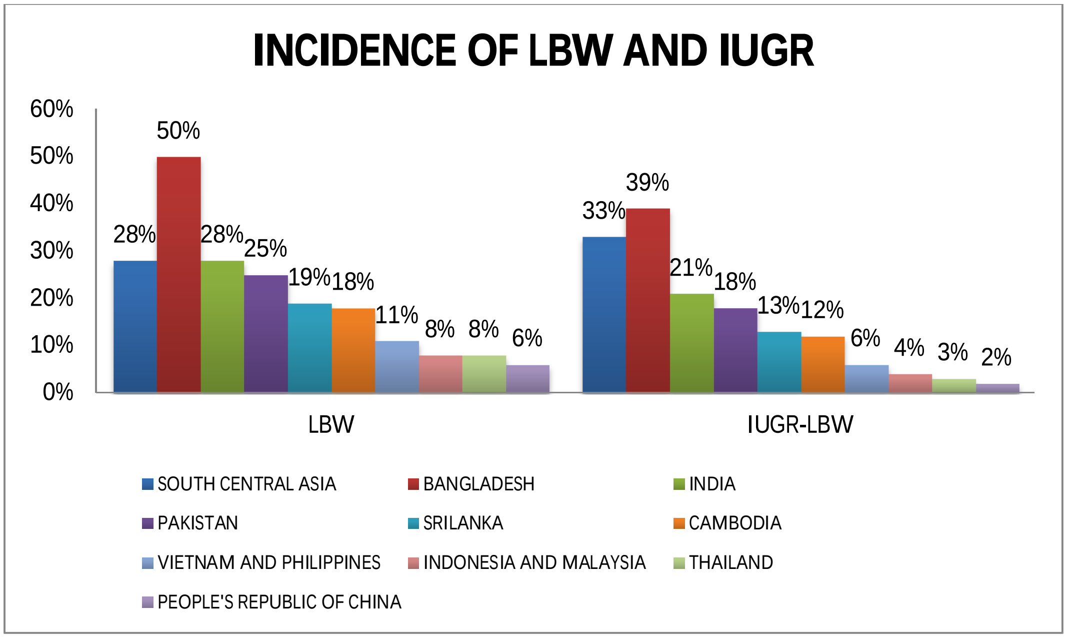


Figure-3: Incidence of Low birth weight (LBW) and IUGR,
as seen in countries of South Central Asia.

A comparison of these statistics to more developed countries, such as the United States, shows that the incidence of SGA births in developed countries is about 10%. One third of these cases represent true IUGR.

Aim of the study

AIM OF THE STUDY

To study the role of L-Arginine supplementation in IUGR complicating pregnancies and its effect on foetal outcome.

FOETAL GROWTH AND DEVELOPMENT

Human foetal growth is characterized by sequential patterns of tissue and organ growth, differentiation, and maturation. Foetal growth has been divided into three phases.

- THE INITIAL PHASE : Phase of hyperplasia occuring in the first 16 weeks and characterized by a rapid increase in cell number.
- THE SECOND PHASE : Extending up to 32 weeks gestation, it includes both cellular hyperplasia and hypertrophy.
- THE THIRD PHASE : Phase of cellular hypertrophy occurring after 32 weeks. Most of the foetal fat and glycogen are accumulated. However there is considerable biological variation in the velocity of foetal growth.

Initial phase	**5 g/day**	**15 weeks gestation**
Second phase	15 to 20 g/day	24 weeks
Third phase	30 to 35 g/day	34 weeks

Table – 1 : The corresponding foetal-growth rates during these three phases

Foetal development is determined by maternal provision of substrate, placental transfer of these substrates, and foetal growth potential governed by the genome. However, the precise cellular and molecular mechanisms by which normal foetal growth occurs are incompletely understood. There is considerable evidence that insulin and insulin-like growth factors, particularly insulin-like growth factor-I (IGF-I), have an important role in regulation of foetal growth and weight gain. These growth factors are produced by virtually all foetal organs and are potent stimulators of cell division and differentiation.

Other hormones implicated in foetal growth have been identified in recent years, particularly hormones derived from adipose tissue. These hormones are known broadly as adipokines and include leptin, the protein product of the obesity gene. Other adipokines under investigation include adiponectin, ghrelin, follistatin, resistin, visfatin, vaspin, omentin-1, apelin, and chemerin.

Foetal growth is also dependent on an adequate supply of nutrients. Glucose transfer has been extensively studied during pregnancy. Both excessive and diminished maternal glucose availability affect foetal growth. Reduction in the maternal glucose levels may result in a lower birth weight. It results only after long-term severe maternal caloric deprivation .

Amino acids undergo active transport from maternal blood to the foetus, which explains the normally higher foetal concentrations. This concentration differential is decreased in growth restriction because of lower foetal amino acid levels and higher maternal amino acid concentrations. The etiology of this altered ratio is uncertain, but there are multiple points at which dysregulation can occur.

DEFINITION OF IUGR

Foetuses with estimated foetal weight less than 10^{th} percentile for that gestational age or two standard deviations below the population mean are considered growth restricted(RCOG 2014)

INCIDENCE

With a prevalence of 3-7% in general population, IUGR can complicate 10% to 15% of all pregnancies. IUGR represents the second most common cause of perinatal mortality, after prematurity.

MEASURE OF MALNOURISHMENT IN SGA BABIES

1. **PONDERAL INDEX (PI)**

It is used to determine the degree of foetal malnutrition. It is defined as the ratio of body weight to length expressed as

$$\mathbf{PONDERAL\ INDEX} = \mathbf{100} \times \frac{\mathbf{WEIGHT(in\ gms)}}{\mathbf{LENGTH(in\ cms)^3}}$$

- Normal value is 2 – 2.5.
- PI of less than 10 percentile reflects foetal malnutrition.
- PI of less than 3 percentile indicates severe foetal wasting.

- In term infants, PI less than 2.2 and Mid Arm/Head Circumference (MAC/HC) less than 0.27 are also considered as features of foetal malnutrition.

2. CLINICAL ASSESSMENT OF NUTRITION SCORE(CAN) SCORE

Metcoff J developed a scoring system, CAN score, for the assessment of nutritional status of the newborns at birth. It includes 9 parameters namely hair, cheeks, neck and chin, arms, legs, back, buttocks, chest and abdomen. A neonate with CAN score of less than 25 is considered malnourished .

3. CEPHALIZATION INDEX

Harel et al. coined the term cephalization index, a ratio of head circumference to body weight. They postulated that higher the brain : body ratio, the more severe is the IUGR.

CLASSIFICATION OF IUGR

There are predominently three types of IUGR.

- Asymmetrical IUGR (malnourished babies)
- Symmetrical IUGR (hypoplastic small for date)
- Mixed IUGR (usually seen in developing countries)

TYPE 1/ SYMMETRICAL /INTRINSIC (20-30%):

It occurs as a result of growth inhibition early in pregnancy i.e. the hyperplastic stage. Any pathological insult at this phase leads to reduced number of cells in foetus and overall decreased growth potential.

Infants with symmetric growth restriction have reductions in all the parameters including weight, length and the head circumference. They are proportionally small. In such cases there will be less than 3 cm difference between the head and the chest circumference.

HC, AC, FL & weight fall below 10^{th} percentile for GA. Hence Ponderal Index is normal (more than 2). Causative factor is usually not correctable.

Causes are: Intrauterine infections (TORCH), chromosomal disorders, congenital malformations.

IUGR according to the time of insult		
Type I	<28 weeks	Symmetric
Type II	30 weeks	Asymmetric
Type III	36 weeks	Postmature

Table – 2 : Classification of IUGR

TYPE 2/ ASYMMETRICAL /EXTRINSIC(70-80%):

It is the most common form of IUGR. Abnormal growth typically begins in the late second or third trimesters, usually after 28 wks of GA. It affects the hypertrophic stage of foetal growth. It occurs as a result of restriction of nutrient supply in utero i.e. uteroplacental insufficiency.

It also causes reduction in the weight and length of the foetus. The foetuses of this type have disproportionately lagging abdominal growth. Foetal abdominal circumference which reflects liver size is greatly reduced. This type of somatic growth restriction occurs due to preferential shunting of oxygen and nutrients to the brain. This allows normal brain and head growth, that is called as “brain sparing effect”. Because of brain-sparing effects, asymmetrical foetuses are protected from the full effects of growth restriction.

This type of growth restriction leads to decreased amniotic fluid, chronic hypoxia and may result in foetal death.

- Maternal factors low pre–pregnancy weight, under nutrition, medical conditions like pre-eclampsia, autoimmune disease, thrombophilias, renal disease, diabetes and essential hypertension, etc.
- HC & FL are normal, AC is decreased, hence Ponderal index is low (less than 2).

It is a correctable form of IUGR, if identified and intervened earlier.

TYPE 3/ INTERMEDIATE IUGR (5-10%):

It is a combination of both the types. Foetal growth restriction occurs during intermediate phase of growth affecting both hyperplasia & hypertrophy. Hence there is a decrease in the number of cell and cell size.

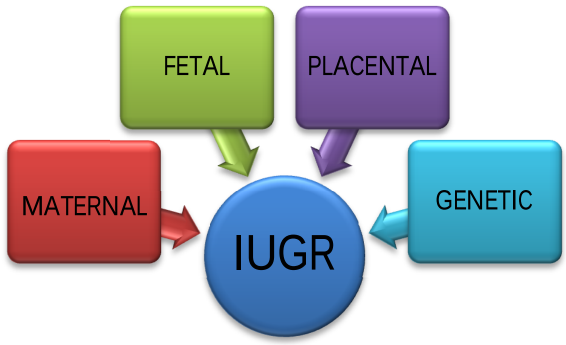
It occurs mostly when early IUGR is affected further by placental causes in late pregnancy. It represents the clinical features of both symmetrical and asymmetrical IUGR. It is associated with chronic hypertension, Lupus nephritis, vascular disease, etc. in early 2nd trimester.

CHARACTERISTICS	SYMMETRICAL IUGR	ASYMMETRICAL IUGR
Time of insult	Earlier gestation	Later gestation
Incidence	20% to 30%	70% to 80%
Etiology	Genetic disorder or infection intrinsic to foetus	Utero-placental insufficiency
Antenatal scan HC, AC, FL, BPD	All are proportionally reduced	AC - decreased BPD,HC,FL- normal
Cell number	Reduced	Normal
Cell size	Normal	Reduced
Ponderal index	Normal (more than 2)	Low (less than 2)
Postnatal anthropometry Weight, length and head circumference.	Reductions in all parameters	Reduction in weight Length and Head circumference- normal (Brain sparing growth)
Features of malnutrition	Less pronounced	More pronounced
Prognosis	Poor	Good

Table 3 : difference between symmetric and asymmetric IUGR

CAUSES

Intrauterine growth restriction may be caused by maternal, placental, or foetal factors. Nearly one-third of IUGRs are due to genetic causes, and two-thirds are related to the foetal environment.



MATERNAL FACTORS :

- Age of the mother (less than 16 years and more than 35 years)
- Maternal hypoxia and high altitude
- Developing country and low socioeconomic status
- Race or ethnicity
- Substance abuse
- Mother on medications like warfarin, steroids, anticonvulsants, antineoplastic, anti-metabolite, and folic acid antagonists.
- Moderate to heavy physical work
- Pre-pregnancy height and weight of the mother (BMI less than 20, weight less than 45 kg and more than 75 kg)
- Parity (more than 5 births)
- Inter pregnancy interval(less than 6 months or 120 months or more)

- Previous SGA birth
- Conception by Assisted reproductive technologies (ART)
- Severe maternal starvation.
- Poor weight gain during pregnancy
- Hematologic and immunologic disorders (Acquired thrombophilias, such as anti-cardiolipin antibodies and lupus anticoagulant)
- Maternal medical disorders like hypertensive disorders, diabetes associated with vasculopathy, chronic renal disease, systemic lupus erythematosus, antiphospholipid syndrome, etc.
- Pathological conditions during pregnancy like preeclampsia and gestational diabetes.
- Maternal infection and parasite infestations (TORCH, malaria, tuberculosis, urinary tract infections and bacterial vaginosis)

PLACENTAL FACTORS

- Placental weight less than 350 grams
- Abnormal utero-placental vasculature
- Placental dysfunction (PIH, pre-eclampsia)
- Thrombophilia-related uteroplacental pathology
- Multiple infarctions
- Partial molar pregnancy
- Single umbilical artery
- Abruptio placenta
- Placental hemangioma

- Placental infections (Placental malaria)
- Chronic villitis of unknown etiology
- Reduced expression of enzymes for redox regulation (thioredoxin, glutaredoxin)

FOETAL FACTORS :

- Constitutional small (50–70% of SGA foetuses, with foetal growth appropriate for maternal size and ethnicity)
- Chromosomal abnormalities [(Trisomies 13, 18, 21), autosomal deletions, ring chromosomes and uniparental disomy]
- Genetic syndromes (Bloom syndrome, Russell-Silver syndrome, Cornelia de Lange syndrome, Dubowitz syndrome, Seckel syndrome, Fanconi syndrome, Roberts syndrome)
- Major congenital anomalies (Tracheo-esophageal fistula, congenital heart disease, congenital diaphragmatic hernia, abdominal wall defects such as omphalocele and gastrochisis, neural tube defect like anencephaly and anorectal malformation)
- Multiple gestation
- Congenital infections (TORCH, Malaria, congenital HIV infection, Syphilis)
- Metabolic disorders (agenesis of pancreas, congenital absence of islets of Langerhans, congenital lipodystrophy, galactosemia, foetal phenylketonuria, transient neonatal diabetes mellitus)

GENETIC FACTORS :

Polymorphisms in maternal, placental and foetal genes encoding for proteins and hormones have been shown to affect the foetal growth. Some of the known genetic associations of intra-uterine growth restriction are

- **PLACENTAL GENES:** Homeobox genes under-expression, NEAT1 (Nuclear Paraspeckle Assembly Transcript 1) over-expression, Placental growth factor (PlGF) under-expression, Trophoblastic miRNAs over-expression, Anti-apoptosis BCL-2 under-expression, Placental Insulin-like growth factor 1 (IGF1) under-expression, Placental Insulin-like growth factor 2 (IGF2) over-expression, Insulin like growth factor binding protein (IGFBP)-3 over-expression, Epidermal growth factor (EGF) under-expression.

- **MATERNAL GENES:** Endothelin-1 (ET-1) over-expression, Leptin under-expression, Visfatin over-expression, Thrombophilia genes (factor V G1691 A or factor IIA(20210)) mutation.

- **FOETAL GENES:** High urinary Protein S100B, Genetic deletion of IGF1 (Insulin Like growth factor 1), Insulin-like growth factors 1 receptor (IGF-1R) mutation.

ENDOCRINE BASIS OF IUGR

The foetal growth depends on various hormones namely insulin, thyroid, adrenal hormones, and pituitary hormones. These hormones promote the growth and development of the foetus and any disruption in these hormonal levels leads to IUGR.

- **INSULIN:** Insulin controls the cell number because it has direct mitogenic effects on cellular development, thereby leading to glucose uptake and consumption by various body tissues and it also decreases protein breakdown. Foetal insulin acts as a signal of availability of nutrients for growth and insulin deficiency leads to IUGR because of reduced uptake and utilization of nutrients.
- **INSULIN-LIKE GROWTH FACTOR:** Insulin-like growth factor-I (IGF-I) is regulated positively by glucose supply in the foetus. It has mitogenic properties causing somatic cell growth and proliferation. It influences the transport of glucose and amino acids across the placenta.
- **THYROID HORMONES:** Foetal hypothyroxinaemia leads to developmental abnormalities such as decreased oxygen consumption and oxidation of glucose, leading to decreased foetal energy supply for growth. It also lowers the circulating and tissue concentrations of IGF-I.
- **PAPP-A:** Pregnancy-associated plasma protein-A (PAPP-A) is secreted by the decidua into the maternal circulation. The function of PAPP-A is to cleave IGFBP-4, a potent inhibitor of IGF action, leading to increase in the activity of local IGFs. The low circulating levels of PAPP-A in early pregnancy have shown to be associated with an increased risk for IUGR.
- **GROWTH HORMONE :** Growth hormone alters maternal nutrient favouring delivery of nutrients to the foetus.

MOLECULAR BASIS OF IUGR

IUGR primarily occurs due to placental insufficiency because of placental ischemia, caused by an imbalance between vasopressors & vasodialators. Vasopressors are thromboxanes (TXA2), angiotensin II and endothelin 1 whereas prostacyclins (PGI2) and nitric oxide act as vasodilators.

Nitric oxide (NO) is synthesized in the vascular endothelium and syncytiotrophoblast from L – Arginine. It is constitutively produced in human vein umbilical cells and platelets from conversion of L-Arginine to L-Citrulline by endothelial NO synthase(eNOS). It causes cyclic guanosine monophosphate (cGMP) mediated vascular smooth muscle relaxation, inhibits platelet aggregation, prevents intravillous thrombosis and thus causes an increase in the foetal blood supply by improving the uteroplacental circulation. Impaired Arginine transport into endothelial cells was observed in the umbilical endothelium from IUGR infants in a clinical study.

It is not known whether an improvement of endogenous NO production could enhance foetal growth. NO-induced vasodilation in renal vessels may improve glomerular filtration rate (GFR) and thereby enhance foetal urine production.

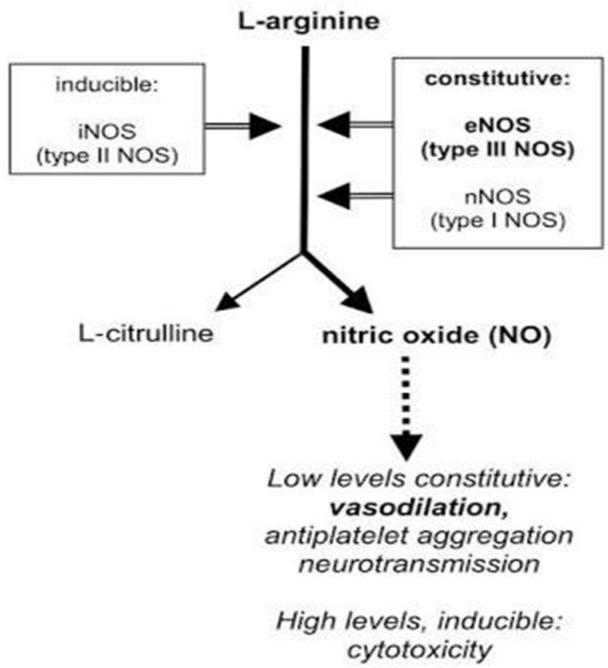


Figure 3: Production of NO from L-Arginine

PLACENTAL TRANSFER OF AMINO ACIDS TO THE FOETUS

Amino acid flux across the trophoblast depends on several factors. Transporter abundance, activity, and affinity as well as villous and microvillous surface area all affect transport capacity. Transport properties change as gestation advances to augment total amino acid exchange capacity and support exponential foetal growth. These properties are affected during an IUGR pregnancy.

The maternal plasma amino acid profile is a major factor in determining protein delivery to the foetus. While maternal diet plays a key role in determining concentrations of maternal amino acids, maternal body composition (lean body mass) and maternal protein turnover and metabolism also affect circulating amino acids.

AMINO ACIDS AND IUGR

Human studies have documented decreased concentrations of certain amino acids including the BCAA, threonine, and Arginine, while others have not found differences. While amino acid concentrations in IUGR foetuses are variable, a consistent feature in both human and animal studies is reduced placental transfer of certain essential amino acids. Furthermore, the severity of IUGR correlates with the severity of decreased amino acid transfer.

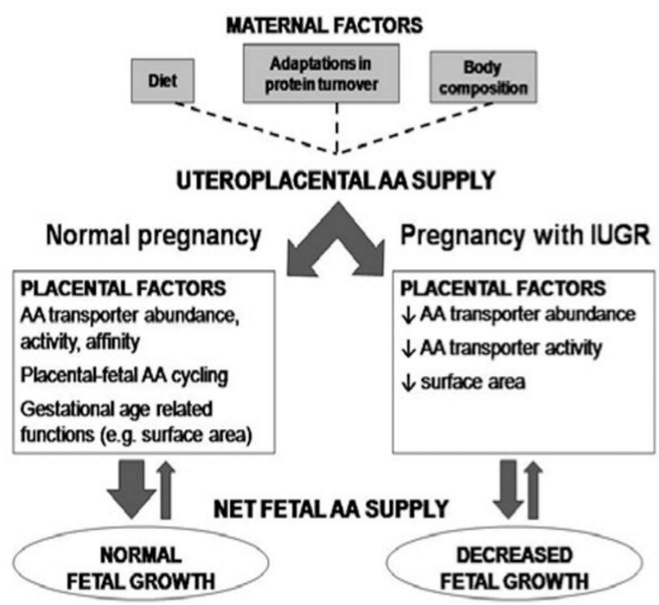


Figure 4 : Determinants of net fetal amino acid (AA) supply.

ROLE OF L-ARGININE IN PREGNANCY AND FOETAL GROWTH

L-Arginine is a versatile amino acid with a wide range of biological functions. The "L" in the name refers to the left-handed configuration of the molecule.It serves as a precursor not only to proteins but also nitric oxide which has been identified as endothelium-derived relaxing factor.

There are several proposed mechanisms by which Arginine supplementation might improve foetal growth.

a. Increasing uteroplacental perfusion and foetal nutrient delivery by increasing local nitric oxide (NO) concentrations.
b. A second mechanism is Arginine mediated stimulation of maternal growth hormone secretion.
c. A third potential mechanism is enhancement of placental growth and development via the promotion of polyamine synthesis.
d. Arginine, in modest to high amounts, is a potent foetal insulin secretagogue, and insulin is a major anabolic hormone in the foetus.
e. Finally, Arginine has been shown to stimulate skeletal muscle protein synthesis.

FDA CATEGORY :

It is a category B drug.

RECOMMENDED DOSAGE

L-Arginine has been studied at oral doses of 6 to 30 g/day for a variety of conditions. Many formulations have been used.

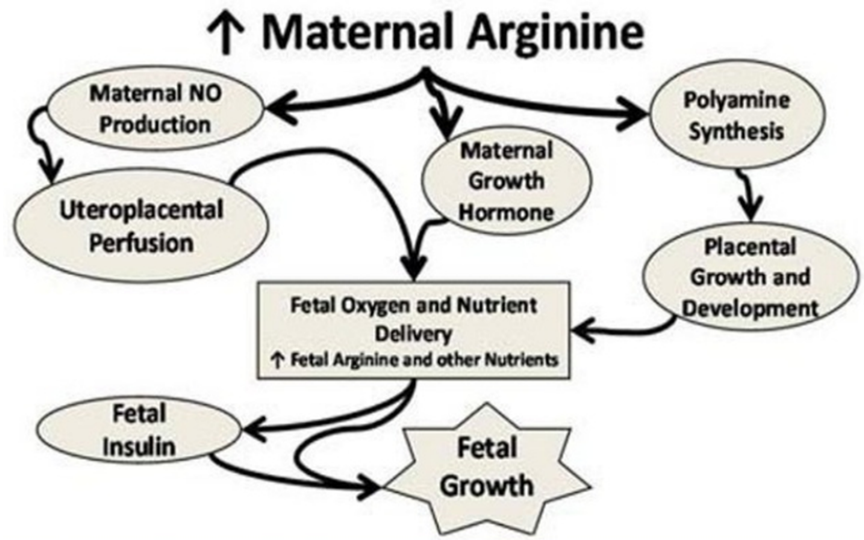


Figure 5 : Mechanisms by which Arginine might improve fetal growth

HOW SAFE IS IT?

CONTRAINDICATIONS

Absolute contraindications have not been identified. L-Arginine is not recommended in patients following an acute heart attack.

PREGNANCY/NURSING

Specific information regarding safety and efficacy during pregnancy and breastfeeding is lacking, although several trials have been conducted in pregnant women without notable ill effects.

INTERACTIONS

L-Arginine has unpredictable effects on insulin and cholesterol lowering agents. L-Arginine may increase the effects of isosorbide mononitrate and other nitric oxide donors, such as glyceryl trinitrate and sodium nitroprusside.

SIDE EFFECTS

L-Arginine has few reported adverse reactions. Nausea and diarrhea have been reported infrequently. Bitter taste may occur with higher doses. Due to its ability to dilate blood vessels, low blood pressure may occur.

Dera et al. in a study in 2007 which included 69 randomly chosen pregnant women diagnosed with gestational hypertension or whose fetuses were diagnosed with intrauterine growth restriction had 42 women receive 3 g L-Arginine daily as a supplement to standard therapy and 27 women receive placebo as well as routine therapy. Use of L-Arginine was associated with lower rate of operative deliveries and higher APGAR scores at both 1 and 5 minutes. While their study demonstrated that L-Arginine administration to pregnant women with gestational hypertension and growth restriction may improve fetal condition and neonatal outcome after delivery by prolonging pregnancy, the authors concluded that these benefits required confirmation by a larger, more powered study.

MOLECULAR BASIS OF L-ARGININE DEFICIENCY IN IUGR

Morris et al., reported that gene expression and protein tissue content of arginase II (enzyme that degrades Arginine to orthinine) were found to be higher in preeclamptic villi than in normotensive pregnancy. Therefore a lower than normal L-Arginine concentration caused by arginase II over expression redirects endothelial isoform of nitric oxide synthase towards peroxynitrite.

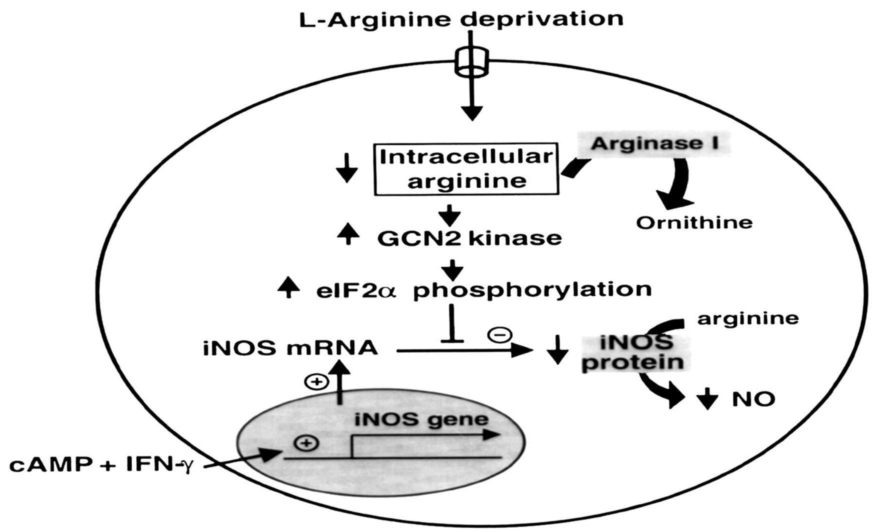


Figure 6 - L-Arginine deficiency caused by arginase II over expression

The interaction of NO and superoxide produces peroxynitrate anion, a strong long lived oxidant with pronounced deleterious effects that causes vascular damage. Peroxynitrite is a cytotoxic anion that inhibits mitochondrial electron transport, oxidizes proteins, and initiates lipid peroxidation and nitrates aromatic aminoacid. Peroxynitrite by causing vascular damage contributes to the increased placental vascular resistance.

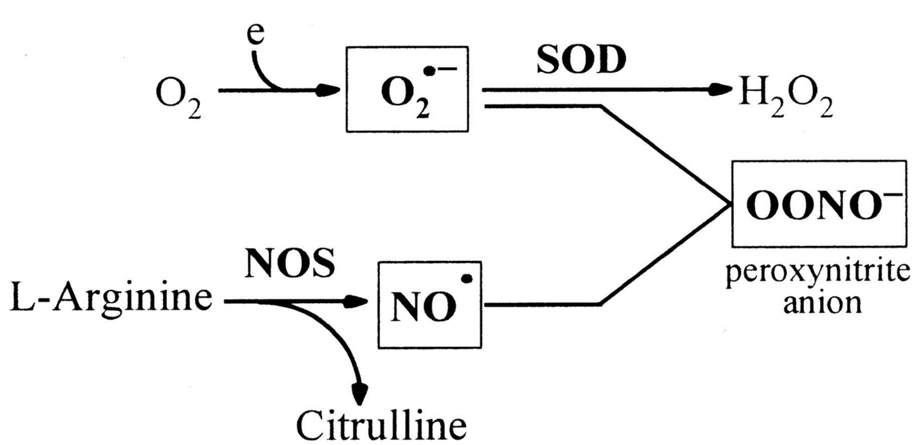


Figure – 7 : Production of peroxynitrite from NO

The combination of a deficiency of NO and increase in peroxynitrite can directly or indirectly initiate a vast majority of physiological & serological changes associated with placental dysfunction, increased thromboxanes and endothelin 1 and decrease in prostacyclins. Therefore, studies recommend the supplementation of L Arginine and antioxidant in pregnancy to maintain the levels of NO so as to facilitate the required vasodilatation and have a beneficial role in the foetal growth.

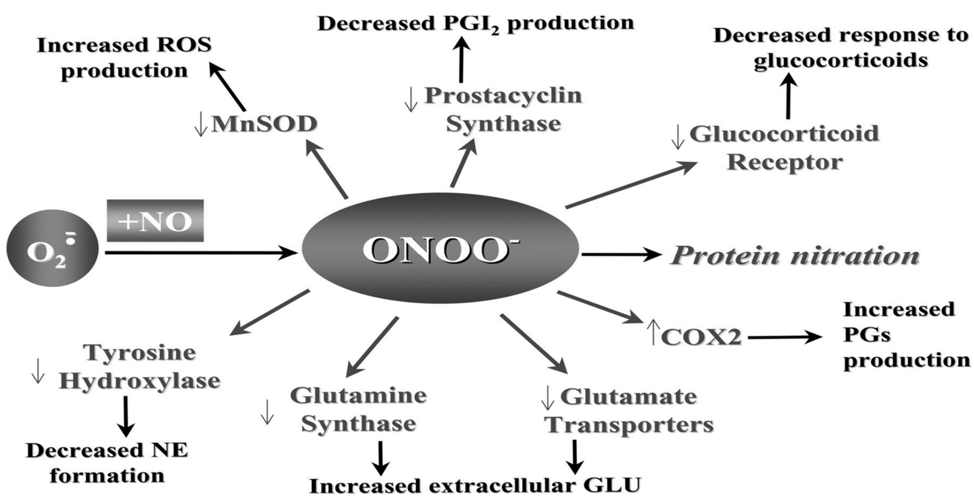


Figure – 8 : Pathophysiology of peroxynitrite effect

CLINICAL FEATURES:

IUGR newborns have typical clinical features. These include

- Weight less than expected for the gestational age
- Relatively large heads for the size of the body in asymmetrical IUGR
- Large anterior fontanelle
- Loss of buccal fat, face has a typical shrunken or “wizened” appearance (Old Man look)

- Small or scaphoid appearing abdomen, thin umbilical cord often stained with meconium
- Decreased skeletal muscle mass and subcutaneous fat tissue with thin and arms and legs.
- Long finger nails.
- Relatively large hands and feet with increased skin creases
- Loose folds of skin in the nape of neck, axilla, inter-scapular area and groins
- Diminished breast bud formation and immature female genitalia due to loss of subcutaneous fat.

All infants with features of IUGR must be examined closely to identify features of chromosomal anomalies, TORCH infections and major malformations.

OUTCOME OF IUGR

IMMEDIATE MORTALITY AND MORBIDITIES

These newborn are faced with many problems after birth. Severely affected IUGR infants, deprived of oxygen and nutrients, may have difficult cardiopulmonary transition with perinatal asphyxia, meconium aspiration, or persistent pulmonary hypertension.

Immediate neonatal complications include asphyxia, hypothermia, hypoglycemia, hyperglycaemia, hypocalcaemia, polycythemia, jaundice, feeding difficulties, feed intolerance, necrotizing enterocolitis, late onset sepsis, pulmonary haemorrhage and intrauterine death.

The risk of neonatal mortality was maximum in babies who were both preterm and SGA in comparison to babies who were either SGA or preterm alone **.**

LONG TERM MORBIDITIES

SGA infants are at risk for impaired growth and neurodevelopment. The behavior performance of SGA babies on day 3, compared to AGA babies, was lower. They also had lower neurobehavioral scores in the areas of habituation, motor system, social-interactive and attention.

LONG TERM PHYSICAL GROWTH

Postnatal growth after IUGR depends in part on the cause of the growth retardation, the postnatal nutritional intake, and the social environment.

- Neonates who are symmetrical IUGR i.e. who have growth restriction in first trimester remain small throughout life.
- Those infants whose intrauterine growth was inhibited late in gestation i.e. asymmetrical IUGR, will have catch-up growth after birth and approach their inherited growth potential when provided with an optimal environment and adequate postnatal caloric intake.
- Preterm SGA children have significantly less height, weight and head circumference compared to the others.

LONG TERM NEURODEVELOPMENTAL OUTCOME

Cognitive and neuro-developmental abnormalities are more common in IUGR infants compared with those who were AGA and born at the same gestational age. Undernutrition that affects head circumference before 26 weeks of pregnancy (symmetrical IUGR) has a greater impact on neurologic function than does asymmetrical IUGR. Neurodevelopmental outcome will be worsened in IUGR who have associated illness such as hypoxic ischemic encephalopathy(HIE) and hypoglycaemia.

DIAGNOSIS

In order to diagnose IUGR, it is essential to estimate gestational age accurately. Although this is usually calculated from the last menstrual period, when known with certainty, the reliability of this estimate is low as timing of ovulation is variable. A first trimester ultrasound can date the pregnancy more reliably.

- According to RCOG green top guidelines, methods employed in the first and second trimesters, to predict the likelihood of a SGA foetus/neonate include: medical and obstetric history and examination, maternal serum screening and uterine artery Doppler.
- The methods of screening for the SGA foetus/neonate in the second and third trimester are abdominal palpation and measurement of symphysis fundal height (SFH) (including customised charts).

1. HISTORY :

- All women should be assessed at booking for risk factors for a SGA foetus/neonate to identify those who require increased surveillance.
- Women who have a major risk factor (Odds Ratio [OR] > 2.0) should be referred for serial ultrasound measurement of foetal size and assessment of wellbeing with umbilical artery Doppler from 26–28 weeks of pregnancy.
- Women who have three or more minor risk factors should be referred for uterine artery Doppler at 20–24 weeks of gestation.

Women that have previously had a SGA neonate have at least a twofold increased risk of a subsequent SGA neonate. The risk is increased further after two SGA births. Women with a prior history of other placenta–mediated diseases are also at increased risk of a subsequent SGA neonate. This includes prior pre-eclampsia and prior stillbirth, and in particular those with a history of previous preterm unexplained stillbirth, due to the association with IUGR.

Maternal medical conditions associated with an increased risk of a SGA neonate are diabetes with vascular disease, moderate and severe renal impairment (especially when associated with hypertension), antiphospholipid syndrome and chronic hypertension. Systemic lupus erythematosus and certain types of congenital heart disease, in particular cyanotic congenital heart disease, are associated with increased likelihood of a SGA neonate.

MINOR RISK FACTORS	MAJOR RISK FACTORS
Maternal age ≥35 years.	Maternal age >40 years.
IVF singleton pregnancy.	Smoker - ≥11 cigarettes per day.
Nulliparity.	Paternal or maternal SGA.
BMI <20.	Cocaine use.
BMI 25-34.9.	Moderate alcohol intake
Smoker - 1-10 cigarettes per day.	Daily vigorous exercise.
Low fruit intake pre-pregnancy.	Previous SGA baby.
Pregnancy interval <6 months.	Previous stillbirth.
Pregnancy interval ≥60 months.	Chronic hypertension.
	Diabetes with vascular disease.
	Renal impairment.
	Anti-phospholipid syndrome.
	Heavy vaginal bleeding during the first trimester.
	Pregnancy associated plasma protein-A (PAPP-A) <0.4 multiples of the median (MOM)

CLINICAL EXAMINATION

1. **SERIAL FUNDAL HEIGHT ESTIMATION**

- Serial fundal height estimation is a simple technique for assessing foetal growth.
- The measurement begins from the fundus till the symphysis pubis with a non-elastic tape with its centimetre side facing-down to avoid manipulation.
- Serial measurement of symphysis fundal height (SFH) is recommended at each antenatal appointment from 24 weeks of pregnancy as this improves prediction of a SGA neonate.
- Women with a single SFH which plots below the 10th centile or serial measurements which demonstrate slow or static growth by crossing centiles should be referred for ultrasound measurement of foetal size.

2. **BIOCHEMICAL MARKERS USED FOR DOWN SYNDROME (DS) SCREENING**

Due to their placental origin, several biochemical markers have been investigated as screening tests for a SGA foetus. A low level (< 0.415 MoM) of the first trimester marker PAPP–A should be considered a major risk factor for delivery of a SGA neonate.

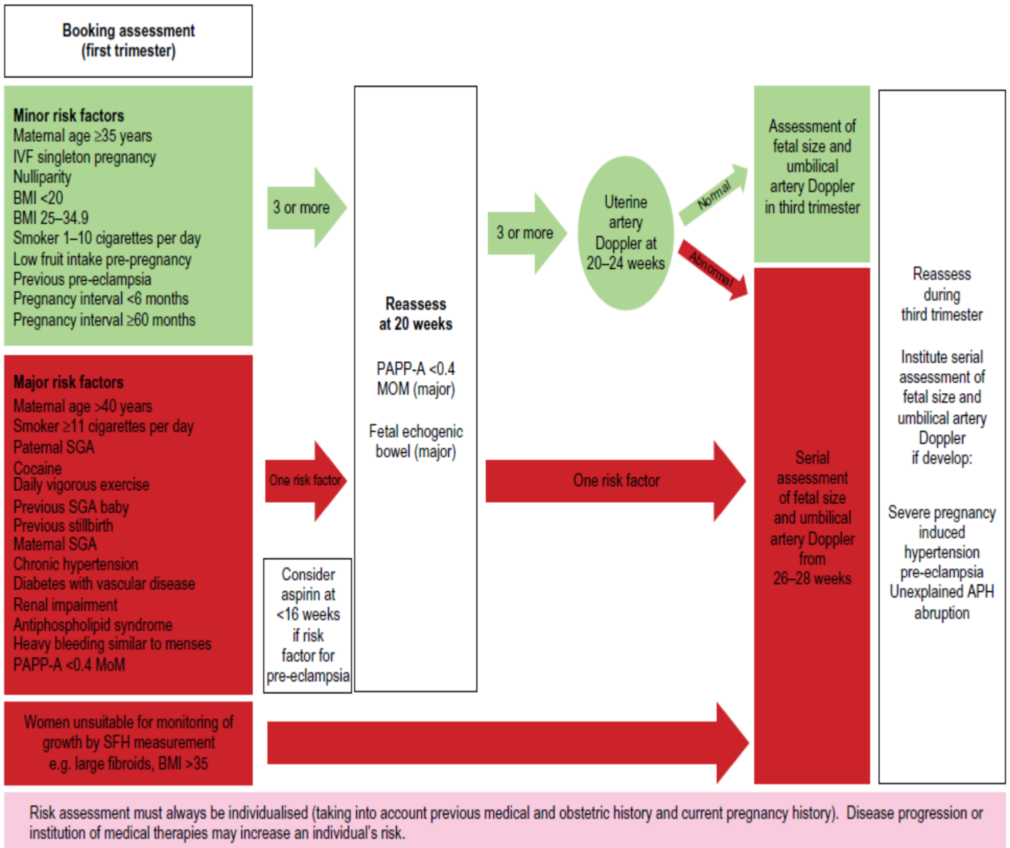


Figure – 9 : Reproduced from: Royal College of Obstetricians and Gynaecologists. The Investigation and Management of the Small–for–Gestational–Age Foetus. Green-top Guideline No. 31. London: RCOG; 2014

3. ULTRASONOGRAPHY

BIOMETRY

- Ultrasound measurements used are biparietal diameter(BPD), head circumference(HC), abdominal circumference(AC) and femur length(FL).
- Measurements below the 10th percentile are highly suspicious of IUGR and measurements below 3rd percentile are unequivocal evidence of IUGR.

- Growth in the abdominal circumference of less than 1 cm over 14 days is also indicative of IUGR.
- When using two measurements of AC or EFW to estimate growth velocity, they should be at least 3 weeks apart to minimise false–positive rates for diagnosing IUGR.
- Reported mean growth rates for AC and EFW after 30 weeks of gestation are 10 mm/14 days and 200 g/14 days. A change in AC of < 5mm over 14 days is suggestive of IUGR.

AMNIOTIC FLUID VOLUME :

- Amniotic fluid is derived from foetal urine and from respiratory tract. In IUGR, shunting of blood from the splanchnic circulation results in reduced renal blood flow, reduced glomerular filtration rate and hence less liquor.
- The amniotic fluid index is measured by adding the vertical depth of cord free amniotic fluid pools in each of the four uterine quadrants. A combined depth of 5 cm or more is normal. Similarly a single deepest vertical pocket(SDVP) of amniotic fluid of more than 2 cm is considered as normal.

DOPPLER ULTRASOUND ASSESSMENT :

- The various Doppler velocities that are being used for assessing fetal wellbeing and detection of IUGR are uterine artery Doppler, umbilical artery Doppler, middle cerebral artery Doppler, cerebro-placental ratio (CPR), and ductus venosus Doppler.

- Uterine arteries provide knowledge of maternal circulation, whereas the umbilical and middle cerebral arteries give information about the fetal circulation.
- The various Doppler abnormalities seen in IUGR are increased resistance in blood vessels or absent and reverse end diastolic flow (AREDF). Increased umbilical artery Doppler perfusion index (PI) has shown good correlation of early identification of IUGR, both alone or else with the cerebro-placental ratio (CPR) ratio. AREDF is usually associated with injury to various fetal organs or death.
- The sequence of abnormal events that herald adverse perinatal outcome begins with an absence of UA end diastolic flow. Later findings include abnormal Doppler pulsatility of the MCA (with decreased PI) and abnormal ductus venosus flow (absent or reversed flow during atrial contraction) and reversed flow in the UA. These changes are significantly associated with perinatal death.

UTERINE ARTERY DOPPLER

- Women with an abnormal uterine artery Doppler at 20–24 weeks (defined as a pulsatility index [PI] > 95th centile) and/or notching should be referred for serial ultrasound measurement of foetal size and assessment of wellbeing with umbilical artery Doppler commencing at 26–28 weeks of pregnancy.
- Women with a normal uterine artery Doppler do not require serial measurement of foetal size and serial assessment of wellbeing with

umbilical artery Doppler unless they develop specific pregnancy complications, for example antepartum haemorrhage or hypertension. However, they should be offered a scan for foetal size and umbilical artery Doppler during the third trimester.

UMBILICAL ARTERY DOPPLER

The umbilical artery (UA) was the first vessel to be studied by Doppler ultrasonography. In the presence of placental insufficiency with progressive severity, there is a higher placental resistance, indicated by a high pulsatility index, absent or reversed end-diastolic velocities of the umbilical artery waveform. Absent or reversed end-diastolic flow velocities in the umbilical arteries are associated with worse perinatal outcome and high perinatal mortality, depending on gestational age.

Doppler flow indices normal : Repeat surveillance every 14 days.

Doppler flow indices abnormal :

- Repeat surveillance twice weekly in foetuses with end–diastolic velocities present
- Daily in foetuses with absent/reversed end–diastolic frequencies.

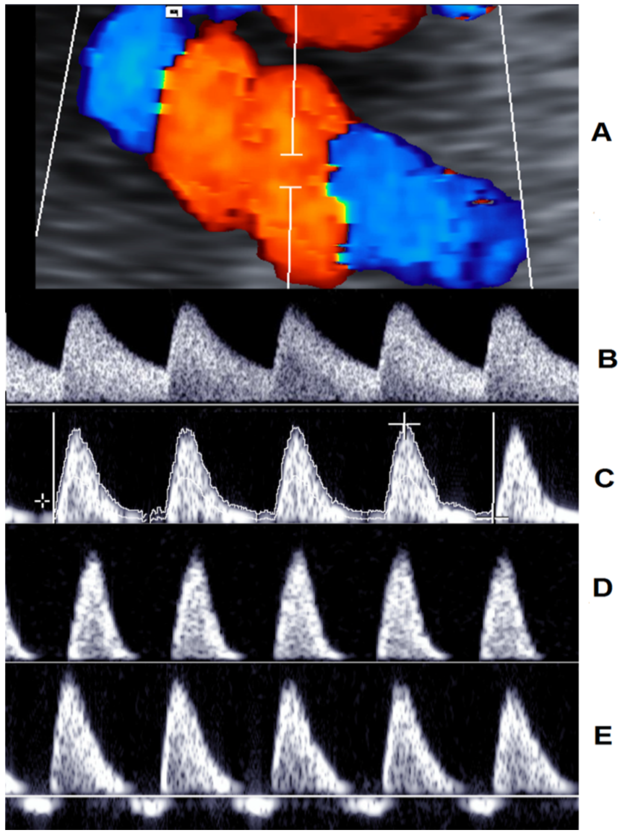


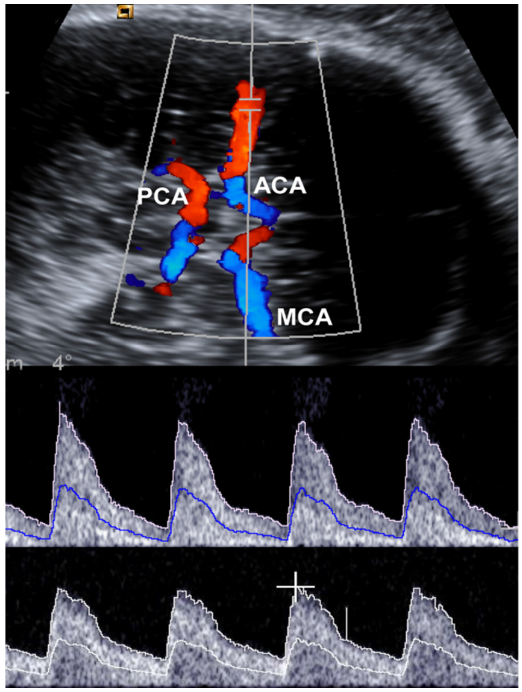
Figure 10 : A, Site of insonation of the umbilical artery Doppler. Progressive waveform patterns with advancing severity were: B, normal umbilical artery waveform, C, increased impedance to flow, D, absent end-diastolic flow, and E, reversed end-diastolic flow.

MIDDLE CEREBRAL ARTERY DOPPLER :

Middle cerebral artery (MCA) is another vessel that has been shown to be affected by IUGR as well. MCA normally exhibits low amplitude of diastolic flow which increases in the presence of foetal hypoxia as a marker of cerebral vasodilation. This most commonly represents a later stage in the hypoxic process and typically occurs after changes in the uterine artery.

Cerebral vasodilatation is a manifestation of the increase in diastolic flow, a sign of the 'brain–sparing effect' of chronic hypoxia, and results in decreases in

Doppler indices of the middle cerebral artery (MCA) such as the PI. Reduced MCA PI or MCA PI/umbilical artery PI (cerebroplacental ratio) is therefore an early sign of foetal hypoxia in SGA foetuses.



A

B

C

Figure 11 : A, Color Doppler assessment of the MCA at the level of the circle of Willis. B, Normal and abnormal (high diastolic velocities and decreased pulsatility index) C, waveforms are shown. *ACA*, anterior cerebral artery; *MCA*, middle cerebral artery; *PCA*, posterior cerebral artery.

DUCTUS VENOSUS (DV) AND UMBILICAL VEIN (UV) DOPPLER :

Ductus venosus doppler has moderate predictive value for acidaemia and adverse outcome. Ductus venosus Doppler should be used for surveillance in the preterm SGA foetus with abnormal umbilical artery Doppler and used to time delivery.

The Ductus venosus (DV) Doppler flow velocity pattern reflects atrial pressure–volume changes during the cardiac cycle. As IUGR worsens velocity reduces in the DV a–wave owing to increased afterload and preload, as well as increased end–diastolic pressure, resulting from the directs effects of hypoxia/acidaemia and increased adrenergic drive. A retrograde a–wave and pulsatile flow in the umbilical vein (UV) signifies the onset of overt foetal cardiac compromise.

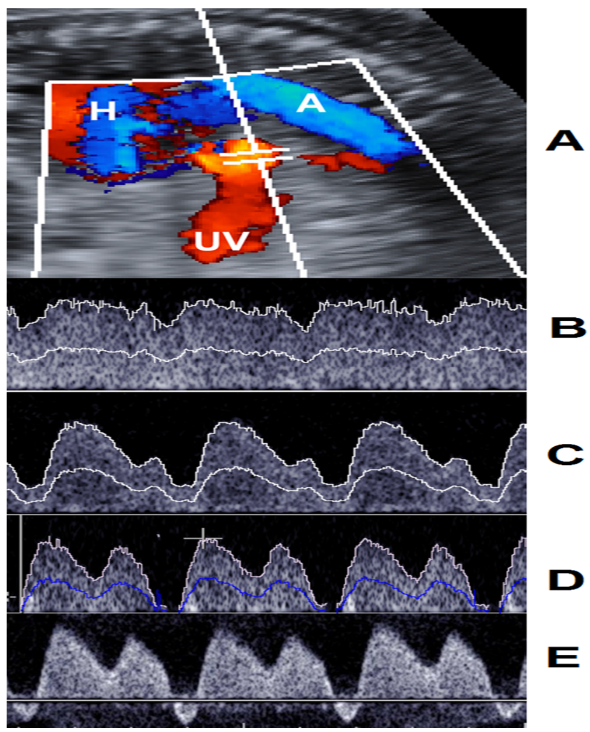


Figure 12 : A, Site of insonation of the DV with color Doppler. Progressive waveform patterns with advancing severity are shown: B, normal DV waveform, C, increased impedance to flow, D, absent enddiastolic flow, and E, reversed end-diastolic flow. *A*, descendent aorta; *DV,* ductus venosus; *H*, heart; *UV,* umbilical vein.

2. INVESTIGATIONS INDICATED IN SGA FOETUSES

- Detailed foetal anatomical survey and uterine artery Doppler by a foetal medicine specialist if severe SGA is identified at the 18–20 week scan.
- Karyotyping in severely SGA foetuses with structural anomalies
- Serological screening for TORCH infection
- Testing for syphilis and malaria in high risk populations.

STAGING OF IUGR

Staging of intrauterine growth-restricted foetuses has been purposed by Mari et al and is based on foetal biometry (expected foetal weight [EFW], abdominal circumference [AC]), Doppler cardiovascular changes, amniotic fluid volume and clinical parameters. This staging is applicable for pregnancies for any gestational age. The classification includes the following:

Stage 0	Foetuses with an EFW or an AC ,10th percentile. Doppler of the UA and MCA is normal.
Stage I	Foetuses whose EFW or AC is ,10th percentile plus abnormal Doppler flow of the UA or MCA.
Stage II	Foetuses whose EFW or AC is ,10th percentile plus absent or reversed Doppler flow of the UA
Stage III	Foetuses whose EFW or AC is ,10th percentile plus absent or reversed Doppler flow of the DV

Based on the amniotic fluid index (AFI), each IUGR foetus will be either A (AFI <5 cm) or B (AFI >5 cm).

MANAGEMENT

The management of IUGR foetus can be

a. Medical management in antenatal period
b. Delivery of the foetus

After identifying a foetus with IUGR, an accurate and thorough history and physical examination are the most important tools for differentiating foetal from maternal and placental causes. The following investigations are used to decide on antepartum management and to decide on delivering the foetus and optimize foetal outcomes:

- Serial measurements of blood flow velocity in foetal vessels using Doppler ultrasound.
- Non-stress test
- Biophysical profile (a composite score based on foetal breathing movements, gross body movements, foetal tone, foetal heart rate, and qualitative amniotic fluid volume)
- Serial foetal weight assessments
- Amniotic fluid volume
- A detailed foetal anatomic survey by ultrasound (as major congenital anomalies are frequently associated with foetal growth restriction).

The guidelines of the Royal college of Obstetrics and Gynaecology (RCOG) recommend the management of these IUGR foetuses including both monitoring and delivery methods. Pregnancy is allowed to continue in those cases where there is no evidence of foetal compromise. If there is any decompensated status of the foetus, pregnancy is then terminated.

ANTEPARTUM FOETAL SURVEILLANCE

Umbilical artery Doppler should be the primary surveillance tool in the SGA foetus, as this has shown to reduce perinatal morbidity and mortality in high-risk population.

- **FOETAL KICK COUNT**

Foetal movements follow a circadian pattern and are expressions of foetal well being. The foetus responds to chronic hypoxia by conserving energy and the subsequent reduction of foetal movements is an adaptive mechanism to reduce oxygen consumption.

- **FHR MONITORING BY CARDIOTOCOGRAPHY (CTG):**

Foetal heart rate (FHR) variation is the most useful predictor of foetal wellbeing in SGA foetuses; a short term variation ≤ 3 ms (within 24 hours of delivery) has been associated with a higher rate of metabolic acidaemia (54.2% versus 10.5%) and early neonatal death (8.3% versus 0.5%).

Foetal heart rate monitoring will show a sequence of changes that correlate with worsening in the foetal situation. The usual order is lack of accelerations, decreased variability, and onset of spontaneous decelerations. These changes are dependant on the severity of foetal compromise and the gestational age of the foetus.

- **BIOPHYSICAL PROFILE**

The BPP is a combination of the observation of the foetal behavior with ultrasound (foetal breathing movements, foetal movements, foetal tone and amniotic fluid volume) and FHR monitoring and is a sensitive test to determine exhaustion of foetal reserve.

Biophysical Variable	**Normal (score=2)**	**Abnormal (score= 0)**
Fetal breathing movements	1 episode FBM of at least 30 s duration in 30 min	Absent FBM or no episode >30 s in 30 min
Fetal movements	3 discrete body/limb movements in 30 min	2 or fewer body/limb movements in 30 min
Fetal tone	1 episode of active extension with return to flexion of fetal limb(s) or trunk. Opening and closing of the hand considered normal tone	Either slow extension with return to partial flexion or movement of limb in full extension Absent fetal movement
Amniotic fluid volume	1 pocket of AF that measures at least 2 cm in 2 perpendicular planes	Either no AF pockets or a pocket<2 cm in 2 perpendicular planes

The biophysical profile (BPP) includes four acute foetal variables (breathing movement, gross body movement, tone and CTG, and amniotic fluid volume each assigned a score of 2 (if normal) or 0 (if abnormal). Reducing BPP score is associated with lower antepartum umbilical venous pH and increasing perinatal mortality. BPP is not recommended for foetal surveillance in the preterm SGA foetus.

MEDICAL MANAGEMENT

A pregnancy which has no signs of hypoxia and good Doppler indices can be continued upto 37 weeks with intense maternal and foetal monitoring (according to RCOG).

- **BED REST**

Bed rest in hospital or at home is widely advised. However the benefits of bed rest must be balanced with the risk of thrombosis.

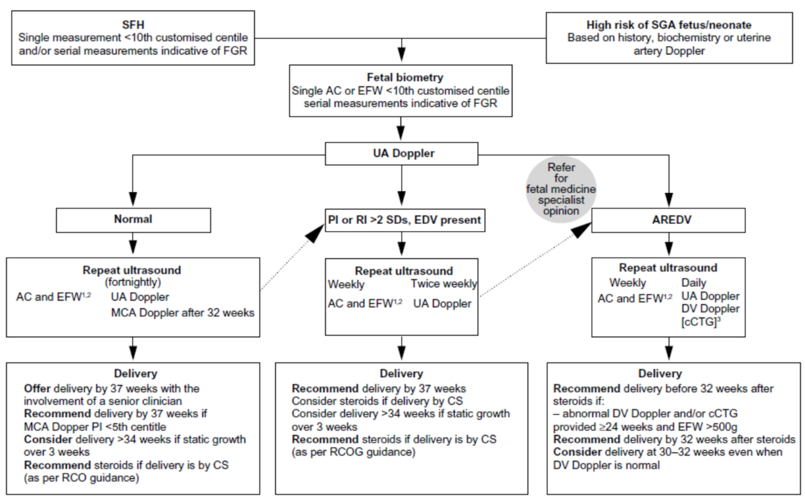


Figure 13 : The Management of the Small–for–Gestational–Age (SGA) Foetus. Reproduced from: Royal College of Obstetricians and Gynaecologists green-top Guideline 2014.

[1]Weekly measurement of foetal size is valuable in predicting birthweight and determining size-for-gestational age.

[2]If two AC/EFW measurements are used to estimate growth, they should be at least 3 weeks apart.

[3]Use cCTGwhen DV Doppler is unavailable or results are inconsistent – recommend delivery if STV <3 ms.

- **MATERNAL DIETARY SUPPLEMENTATION**

Maternal dietary supplementation through balanced caloric intake rather than specific protein supplementation has a variable effect on foetal growth. The effect is small, though foetal weights have been shown to increase by 100–300g.

- **NITRIC OXIDE DONORS**

L-Arginine improves uteroplacental blood flow to overcome placental ischemia by increasing Nitric oxide. This results in vasodilation of uterine arteries. Various studies on effect of NO are as given.

AUTHOR	STUDY	RESULTS
Shalini singh et at	Effect of L-Arginine on Nitric Oxide Levels in Intrauterine Growth Restriction and its Correlation with Fetal Outcome	Mean NO levels were significantly increased and a significant mild reduction in systolic/end-diastolic velocity ratio (S/D ratio) was observed on doppler blood flow study, also neonatal outcome improved and incidences of complications were lowered.
Xiao et al	To investigate the effects of L-Arginine on NO levels and in treating asymmetric fetal growth restriction (IUGR).	Before treatment, mean maternal serum levels of NO2-/NO3- were significantly lower in groups 1 and 2 than in the control group ($P<0.01$). After treatment, maternal serum levels of NO2-/NO3- were considerably higher in group 2 than in group 1 ($P<0.01$). Mean birth weight was significantly higher in group 2 than in group 1 ($P<0.05$), but still lower in group 2 than in the control group ($P<0.01$).

Neri et al.	Effect of L-Arginine on uteroplacental circulation in IUGR in the third trimester	They observed an increase in serum nitrite/nitrate as well as a serum growth hormone levels in every women. They concluded that the subpopulation of IUGR fetuses with impaired uteroplacental circulation, probably the reason of restricted growth, might benefit from L-Arginine supplementation.

- **LOW DOSE ASPIRIN**

Aspirin was effective at lower doses between 50 and 80 mg/day, but the preventive effect was greater at higher doses between 100 and 150 mg/day and among women entered before the 17^{th} week of gestation. Low dose aspirin should not be used routinely in pregnant women.

- **HEPARIN**

Heparin plays an important role in preventing complications of the APLA syndrome. Both heparin and LMWH do not cross the placenta and foetal complications have not been reported. Enoxaparin 40 mg per day subcutaneously or Dalteparin 5000 units per day is injected from the time of confirmation of pregnancy until delivery.

- **SILDENAFIL CITRATE**

Sildenafil citrate, a specific phosphodiesterase inhibitor, is increasingly used for pulmonary hypertension in pregnancy. Sildenafil is also emerging as a potential candidate for the treatment of intra-uterine growth retardation and for premature labour. According to studies conducted, sildenafil citrate vasodilated the feto-placental circulation via a cGMP dependant mechanism involving increased responsiveness to nitrous oxide. Sildenafil growth was associated with increased AC growth (odds ratio 12.9). Randomised controlled trial data are required to determine whether sildenafil improves perinatal outcome for early onset IUGR patients.

DELIVERY OF THE GROWTH RESTRICTED FOETUS

INTRAPARTUM MONITORING

- **FOETAL HEART RATE AUSCULTATION**

Intrapartum surveillance is mainly focused upon detecting foetal asphyxia in order to prevent perinatal death or future neurodevelopmental handicap. Bradycardia, tachycardia and irregular heart rate are signs of asphyxia.

- **ELECTRONIC FOETAL MONITORING**

Uterine contractions of labour reduce the uteroplacental blood flow and/or compress the umbilical cord depending on its position and the quantity of amniotic fluid. Reduction of blood flow may compromise foetuses which have pre-existing hypoxia. The features of reduced baseline variability, decelerations, absence of accelerations suggest hypoxia in the foetus.

- **FOETAL PULSE OXIMETRY**

Foetal pulse oximetry appears to be a promising new tool in intrapartum foetal surveillance. Not only is it accurate and rapid in its measurement of foetal oxygenation, it also affords direct assessment (rather than indirect assessment, as with FHR monitoring) of foetal oxygenation and peripheral tissue perfusion.

- **SCALP BLOOD PH**

This is a useful aid to determine foetal acidosis when difficulties arise with interpretation of abnormal CTG tracings. With CO2 accumulation, due to reduced placental gas exchange, pH decreases as a result of respiratory acidosis. Increasing hypoxia leads to anaerobic metabolism resulting in lactate and hydrogen ion (H?) production.

TIMING OF DELIVERY

- All women with IUGR should be given a course of corticosteroids for fetal lung maturity.
- The timing of delivery in a growth restricted fetus should be guided by both fetal condition and gestational age.
- Early admission is recommended in women in spontaneous labour with a SGA foetus in order to have continuous foetal heart rate monitoring.
- When IUGR is mild and uncomplicated, with normal end-diastolic flow, delivery can be delayed until 37 weeks to allow time for lung maturation.
- Once absent end-diastolic flow in the UA is demonstrated, the biophysical profile (BPP) and Doppler indices should be done twice weekly and daily foetal kick counts is recommended.
- Once reversal of flow in the UA or cephalisation in the MCA is present, hospitalization with continuous oxygen therapy, bed rest, daily BPP and daily

Doppler are indicated. Steroids for foetal lung maturity should be administered.

- A pulsatile pattern in the DV Doppler is highly suggestive of foetal acidemia and is an indication for delivery.

MODE OF DELIVERY :

- In the SGA foetus with umbilical artery AREDV, delivery by caesarean section is recommended.
- In the SGA foetus with normal umbilical artery Doppler or with abnormal umbilical artery PI but end–diastolic velocities present, induction of labour can be offered but rates of emergency caesarean section are increased and continuous foetal heart rate monitoring is recommended from the onset of uterine contractions.

Compared to appropriate–for–gestational age foetuses, term and near term SGA foetuses are at increased risk of FHR decelerations in labour, emergency caesarean section for suspected foetal compromise and metabolic acidaemia at delivery.

NEED FOR THE STUDY

Prevention of low birth weight (LBW) in infants should be recognized as an important public health priority in developing countries, where the condition is largely attributed to IUGR.

High rates of IUGR should be a cause of concern because they not only indicate an imminent risk of malnutrition and morbidity in women of childbearing age but also signal of a high risk of malnutrition, morbidity and mortality for the newborn in the developing countries. Undoubtedly, one of the most important goals of effective antenatal care is the detection of foetuses at risk of suboptimal growth at a stage where there is significant potential for treatment. In India, the problem of IUGR is compounded by poverty and insufficient per capita income.

PREVENTION OF IUGR

Increased incidence of intrauterine growth restriction in developing countries is often due social reasons and does not appear to reduce with interventions targeted at pregnant women alone. Adolescent nutrition, pre-pregnancy weights, poverty, inter-pregnancy interval are the crucial determinants of foetal growth in low and middle income countries. However some interventions exist to improve maternal nutrition and reduce foetal growth restriction and Small-for- Gestational Age (SGA) births in appropriate settings in developing countries, if scaled up before and during pregnancy. The interventions include

a) Balanced energy protein

b) Calcium supplementation

c) Multiple micronutrient supplementations

d) Preventive strategies for malaria in pregnancy

Review of literature

REVIEW OF LITERATURE

This study is based on the articles several research projects on IUGR.

1. Sieroszewski P et al 2004

They conducted a study with two randomly chosen groups of pregnant women with ultrasound-diagnosed intrauterine growth restriction. The study group was treated with L-Arginine and control group without L-Arginine supplementation. The ultrasound estimation of foetal weight at the start and at the end of the treatment showed a significant statistical difference with a mean increase of foetal weight in the study group. There was also a significant difference showing the positive effect of the treatment on the weight of newborns. In the treated group the percentage of growth-retarded newborns was 29% while in the untreated group it was 73%.

2. Mariola ropacka et al 2003-2006

They studied the effectof L-Arginine on foetal outcome in IUGR foetuses. They observed that L-Arginine supplementation accelerated foetal growth. The topic of analysis were: gestational age at delivery, birth weight and mode of delivery, APGAR score in t he 1st and 5thminute, umbilical cord acid-base status, signs of infection, ventricular hemorrhage, respiratory distress syndrome, admission to NICU, placental weight, side effects of current therapy and the appearance IUGR at delivery. Neonates delivered in the L-Arginine group revealed higher APGAR scores, better umbilical cord acid-base status. Moreover, they presented lower incidence of INCH, RDS and admission to NICU. Oral treatment with L-Arginine seems to be promising in improving foetal outcome as well as accelerating foetal growth in pregnancies complicated by IUGR.

3. Shalini Singh et al 2015

They studied the effect of L-Arginine on Nitric Oxide Levels in Intrauterine Growth Restriction and its Correlation with Foetal Outcome. With L-Arginine supplementation, mean NO levels were significantly increased and a significant mild reduction in systolic/end-diastolic velocity ratio (S/D ratio) was observed on doppler blood flow study, also neonatal outcome improved and incidences of complications were lowered. L-Arginine can be used to increase maternal NO levels, enhancing birth weight and decreasing neonatal morbidity.The ideal candidate for Arginine therapy according to their study was IUGR cases with S/D ratio less than 4.96 ± 0.49 and NO levels below 33 lmol/L with minimum of 3 weeks duration of Arginine supplementation.

4. Lampariello C et al 1997

They evaluated the efficiency and use of L-Arginine in asymmetrical intrauterine growth retardation. They were of the conclusion that L-Arginine improves GH-RH incretion, with consequent increase of plasmatic GH influencing somatic growth. L-Arginine moreover, is the obligatory precursor for nitric oxide (NO) enzymatic synthesis. NO helps the prolapse of smooth musculature and, consequently, the improvement of placental blood circulation.

5. Chen et al 2016

They conducted a meta analysis to investigate the effect of L-Arginine and sildenafil citrate on intrauterine growth restriction foetuses. Randomized controlled trials assessing the effects of L-Arginine and sildenafil citrate on IUGR were included in their study. In the L-Arginine treated groups, there was a significant increase in foetal birth weight and gestational age at birth. L-Arginine treated foetuses have a significant reduction in the ratio of neonatal respiratory distress syndrome and intracranial hemorrhage. The results of this meta-analysis showed that L-Arginine increased birth weight and prolonged gestational age at labor of IUGR foetuses.

Materials and methods

MATERIALS AND METHODS

Acknowledging the beneficial effects of L-Arginine on endothelial vasculature, the present study was designed to evaluate the role of L-Arginine in women with IUGR and its correlation with neonatal outcome.

TYPE OF STUDY :

Randomized Control Trial

PERIOD OF STUDY :

March 2017- August 2017

PLACE OF STUDY :

Antenatal OPD, Antenatal Ward, Labour ward, Dept of Obstetrics & Gynaecology, Government Kilpauk Medical College &Hospital, Chennai

STUDY POPULATION :

Pregnant women of gestational ages 28-36 weeks attending Government Kilpauk Medical Hospital.

SAMPLE SIZE :

Sample size was determined based on a hospital based randomised prospective study on correlation between Effect of L-Arginine on Nitric Oxide levels in Intrauterine Growth Restriction and its Correlation with Fetal Outcome authored by Shalini Singh et al, published in IJCB | July-September 2015

DESCRIPTION

- Critical variable of the study is foetal weight increase (1.9kg +/- 0.3kg)
- A confidence interval of 95%
- Standard deviation of 0.3
- Margin of error of 0.08(1.748kg – 2.052kg)

Assuming that 80% as a power of the study, minimum sample size required was 55 (to detect a statistically significant result within a range of 1.748-2.052kg gain in foetal weight). Assuming an attrition rate of 10%, sample size was calculated as 60 subjects.

METHODOLOGY

After obtaining clearance from the hospital ethical committee, this randomized control study was undertaken in the Department of Obstetrics and Gynaecology at Government Kilpauk Medical College and Hospital, Chennai from March 2017 to August 2017. Written informed consent was obtained from the women explaining it to them in their language they best understand. This hospital based study included 60 antenatal women in the age group of 20–35 years, with foetal IUGR (a growth lag of >4weeks clinically or Estimated foetal weight <10th percentile for the GA measured sonographically) selected from antenatal clinics and wards of Kilpauk Medical College and Hospital, Chennai.

The women were divided into two groups. Group I had women receiving L -Arginine therapy (study group) and Group II with women not receiving L -Arginine therapy (control group). Women in group I were supplemented with oral

L-Arginine 3 g once daily for 4 weeks and group II were put on conventional treatment like good high protein diet. Group I and II were matched for age, parity and period of gestation.

GROUPS

GROUPS	DEFINITION	NUMBER
CASES	IUGR women supplemented with oral L-Arginine 3 g once daily for 4 weeks	30
CONTROLS	IUGR women on conventional treatment without L-Arginine	30

INCLUSION CRITERIA :

Women included in this study were those having singleton pregnancies, whose gestational ages during sampling was between 28 and 36 weeks, irrespective of the parity of the patient. Only those women, who had reliable dating of pregnancy confirmed by an early first trimester ultrasound examination using CRL or with known LMP and regular menstrual cycles were enrolled in the study, thereby ruling out the doubt of having wrongly diagnosed IUGR. The study group comprised of women with assymetrical type of IUGR.

EXCLUSION CRITERIA :

Antenatal women where IUGR was a clinical suspicion only and no grey scale ultrasound assessment was done were not included in the study. Women with symmetrical IUGR, women with severe oligohydramnnios with AFI less than 5cms(in the control group as it had ethical issue of not giving L-Arginine), period of gestation less than 28 wks and more than 37 weeks, those having infections like toxoplasmosis, rubella, cytomegalovirus, herpes, syphilis and others (TORCH infections), major maternal medical illness like gestational and overt diabetes mellitus, hypertension (Systemic hypertension and gestational hypertension), severe anemia with haemoglobin(Hb) <8gms%, renal diseases like chronic renal failure, cardiovascular disorder , maternal pulmonary disorder were excluded. Pregnant women with pre-eclampsia, multiple gestation, transverse lie, fetal malformations and congenital anomalies were also excluded. Maternal smoking and history of allergy to food products were also excluded. Pregnant women who had severe placental insufficiency, prelabour rupture of membranes(PROM) and women in labour were not taken into study.

Gestational age was calculated from date of last menstrual period using Naegeles formula in case of reliable dates or by first ultrasound in the first trimester of pregnancy. Detailed history was taken and systemic physical examinations and obstetrics examination were performed on all the women. Symphysio fundal height was measured in centimetres. The measurement was taken from the fundus till the symphysis pubis with a non-elastic tape with its centimetre side facing-down to avoid manipulation. Amniotic fluid index was measured with four quadrant technique which consisted of measuring the largest pool of fluid devoid of cord and foetal parts, found in each of the four quadrants of uterus. The sum of all the measurements constitutes amniotic fluid index (AFI).

After confirming the asymmetric IUGR sonographically, umbilical artery doppler blood flow peak systolic/ end-diastolic velocity ratio (S/D ratio) was done to document abnormality in uteroplacental flow. This vessel normally has forward flow throughout the cardiac cycle and the amount of flow during diastole increases as gestation advances. S/D ratio normally decreases as pregnancy progresses. A high S/D ratio denotes resistance to blood flow to fetus. In extreme cases of growth restriction, end diastolic flow was absent or reversed. The umbilical artery peak S/D ratio was considered abnormal if it was above the 95th percentile for the gestational age.

After getting consent from the patient, blood sample were taken for routine investigations to rule out anaemia, diabetes in the mother. All patients were monitored according to standard procedure. Fetal wellbeing was assessed (clinically and sonographically) before and after the therapy in both study and control group. Foetal movements and foetal heart rates were recorded serially. Foetal kick count and non-stress test was performed as and when indicated and documented. Iron, calcium, multi – vitamins supplements and high protein diet were continued orally in both the groups. These women were followed till delivery. The decision about the time and mode of delivery in each case depended on the conditions of the mother and fetus. As far as fetal well-being was observed, spontaneous delivery was recommended. In case of fetal distress a caesarean section was performed. If the mother or foetuses developed complications anytime during the study, the pregnancy was terminated and the patient excluded from the study.

A predesigned study proforma was filled for each case. Data collected were tabulated and analysed. Outcome variables that were analysed were age of the mother, gestational age at entry into the study, pre-treatment AFI & foetal weight, post-treatment foetal weight and AFI(in Group I), gestational age at delivery, mode of delivery, neonatal APGAR score, birth weight, admission in Neonatal intensive care unit (NICU) and duration of stay in NICU. We also analysed the need for resuscitation at birth and complications developed in the neonate. The mortality and morbidity during pregnancy and after delivery were observed, by measuring the number of live births and neonatal deaths.

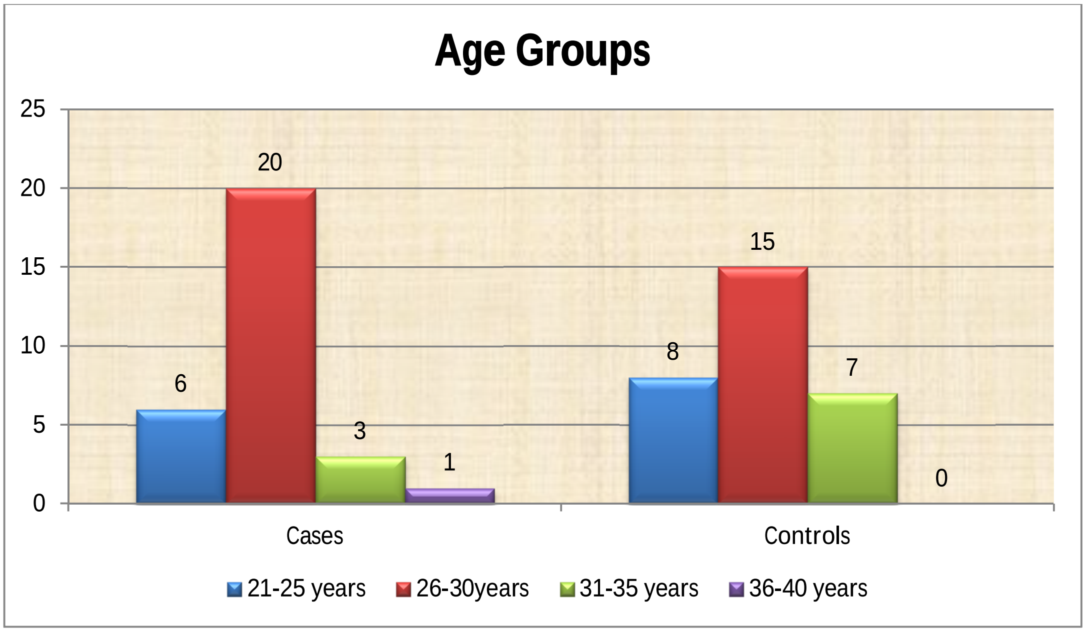
STATISTICAL ANALYSIS:

Descriptive statistics was done for all data and suitable statistical tests of comparison were done. Continuous variables were analysed with the student's t test (paired and unpaired) and categorical variables were analysed with the Chi-Square Test and Fisher Exact Test. Statistical significance was taken as $P < 0.05$. The data was analysed using EpiInfo software (7.1.0.6 version; Center for disease control, USA) and Microsoft Excel 2010.

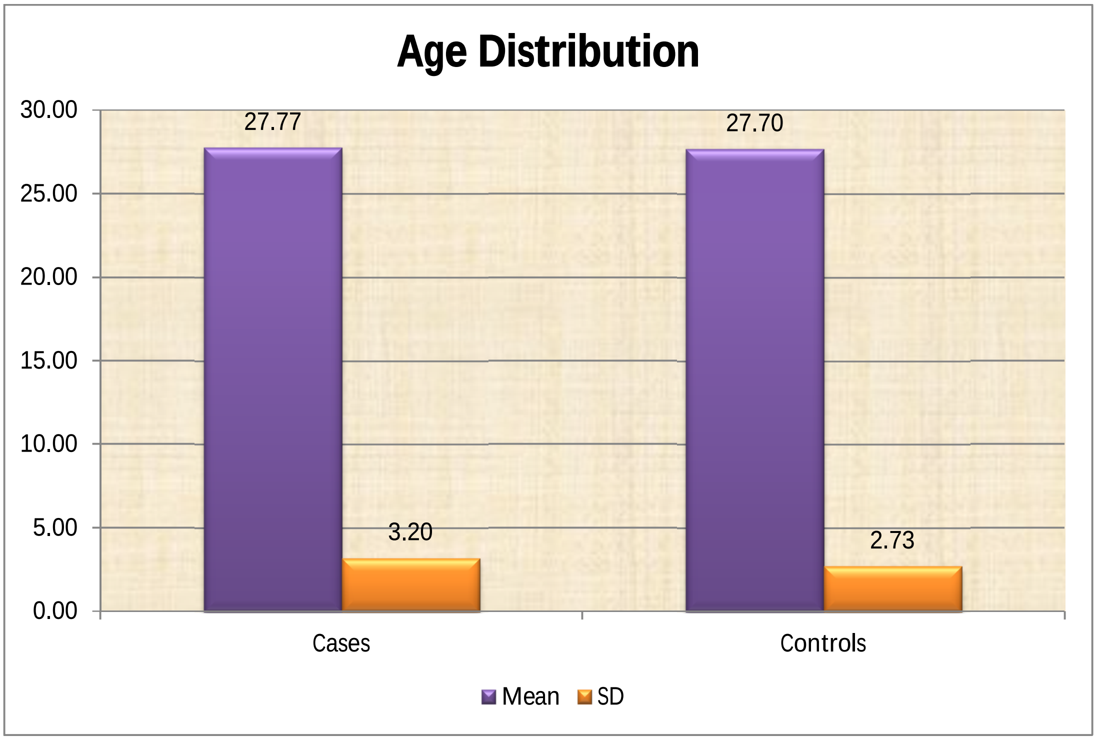
Results

RESULTS

AGE



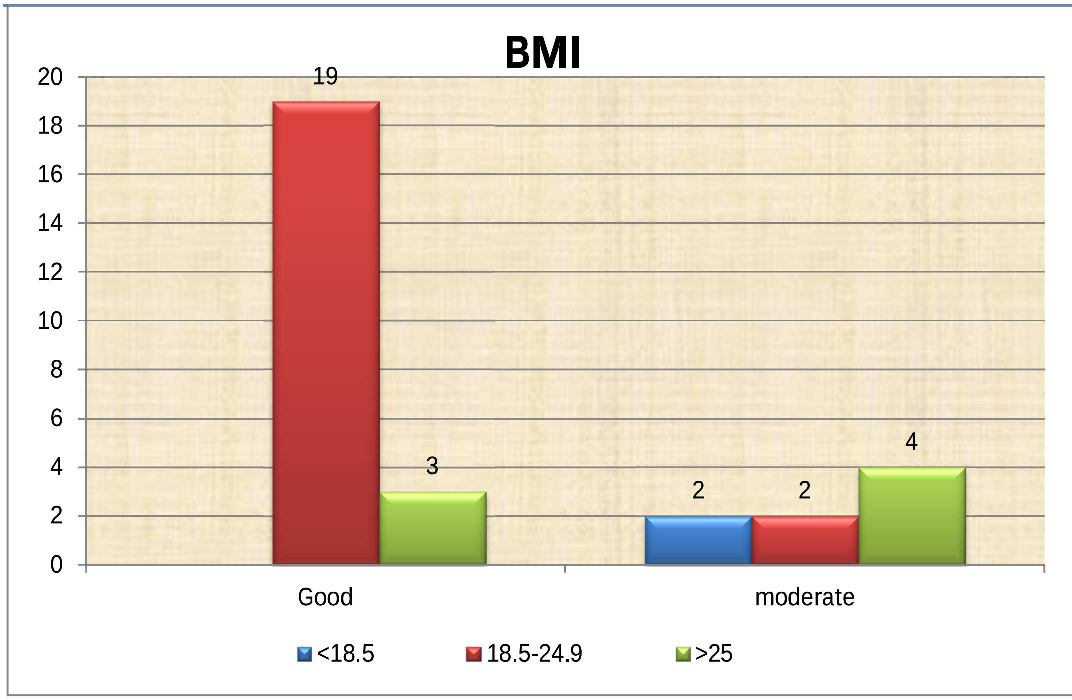
Age Groups	Cases	%	Controls	%
21-25 years	6	20.00	8	26.67
26-30 years	20	66.67	15	50.00
31-35 years	3	10.00	7	23.33
36-40 years	1	3.33	0	0.00
Total	30	100.00	30	100.00



Age Distribution	Cases	Controls
Mean	**27.77**	27.70
SD	**3.20**	2.73
P value (Unpaired t Test)		0.9312

Distribution of ages in both the groups was 26-30years : cases group (n=20, 66.67%) control group (n=15, 50.00%) (p=0.9312, unpaired t test). The difference in the mean age of patients in cases group (27.77) and control group (27.70) was found to be statistically insignificant (p >0.05).

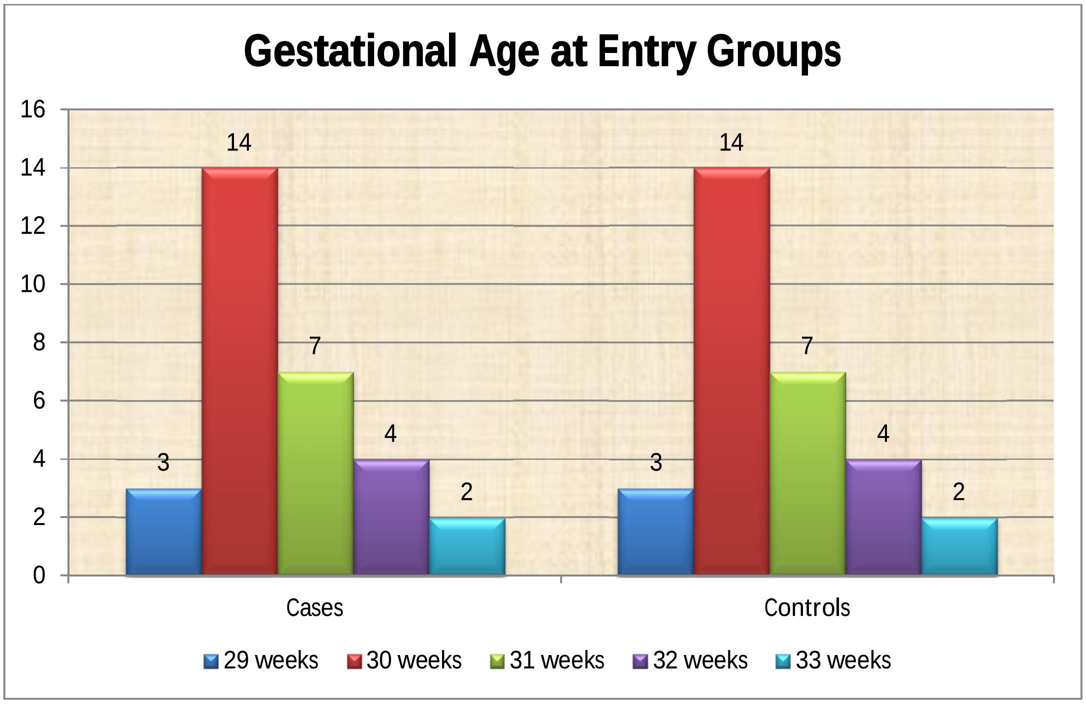
BMI



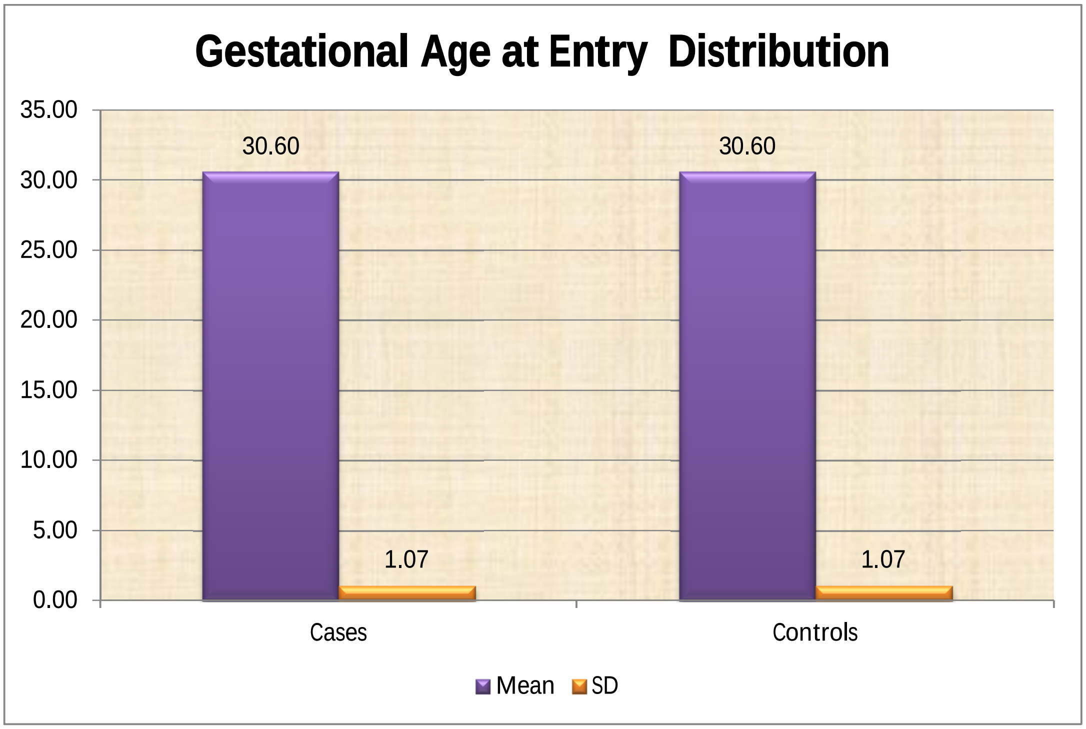
BMI	**No. Of cases**	**Improvement of IUGR status**	
		Good (100%)	**Moderate (80%)**
<18.5	2	0	2
18.5-24.9	21	19	2
>25	7	3	4
P value (Chi squared test)			0.002503

A p value <0.05 indicates the statistical significane of BMI on IUGR improvement.

GESTATIONAL AGE AT ENTRY



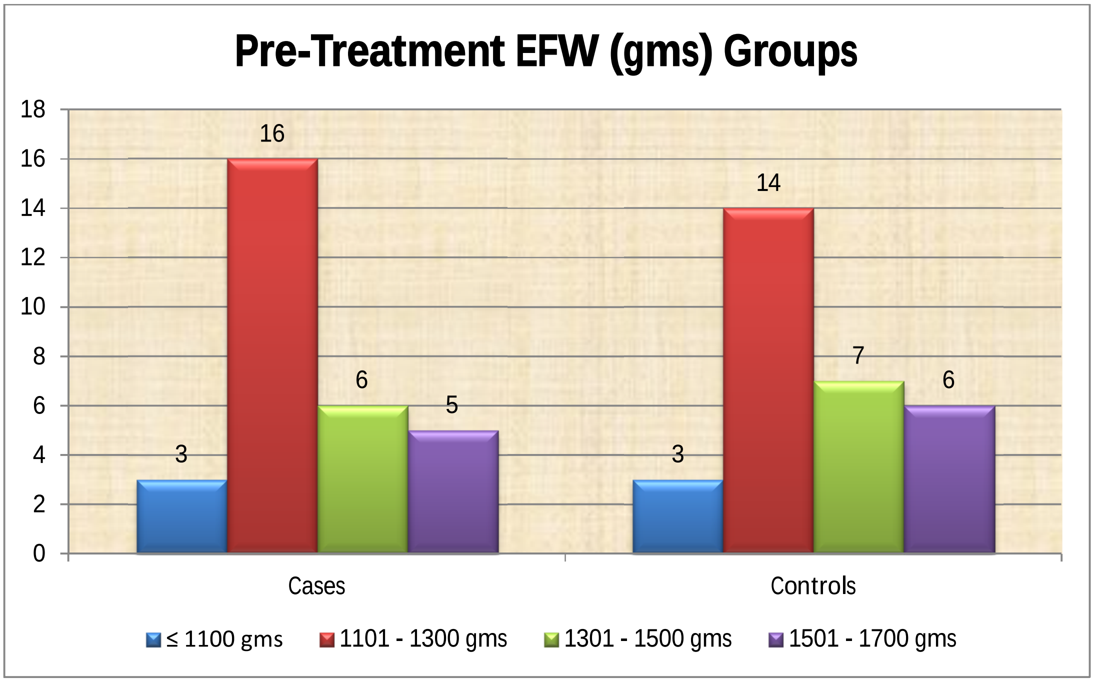
Gestational Age at Entry Groups	Cases	%	Controls	%
29 weeks	3	10.00	3	10.00
30 weeks	14	46.67	14	46.67
31 weeks	7	23.33	7	23.33
32 weeks	4	13.33	4	13.33
33 weeks	2	6.67	2	6.67
Total	30	100.00	30	100.00



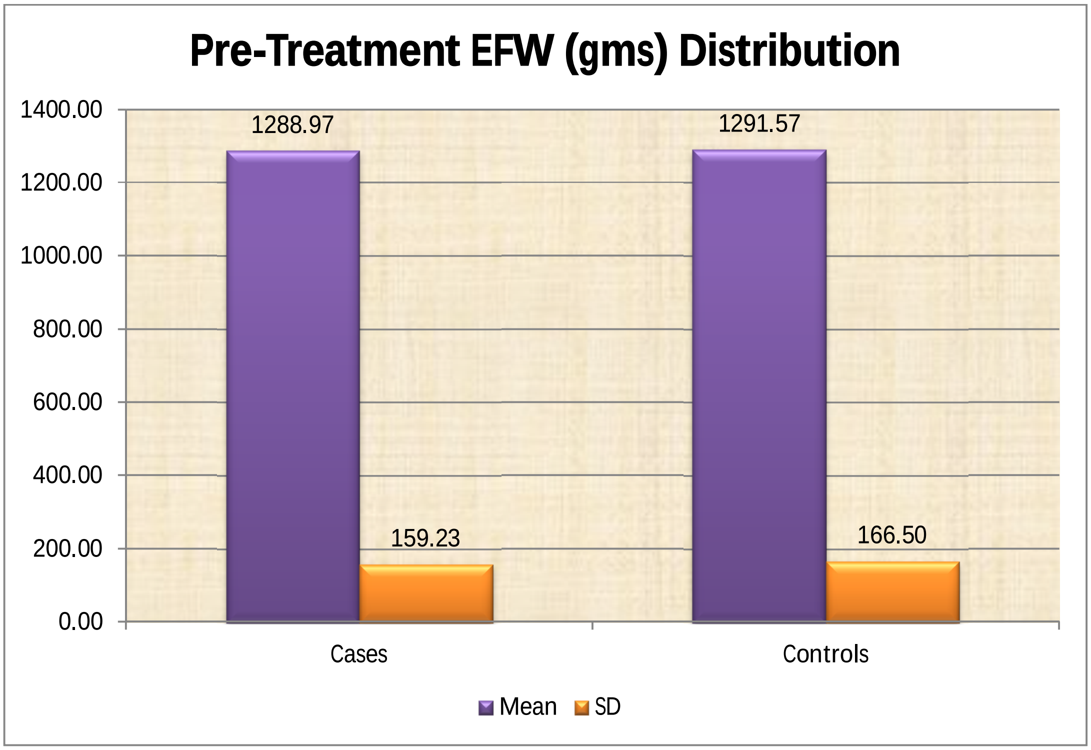
Gestational Age at Entry Distribution	Cases	Controls
Mean	30.60	30.60
SD	1.07	1.07
P value (Unpaired t Test)		>0.9999

In case group(n=14, 46.67%) and control group(n=14, 46.67%), most of the patients were distributed in 30 weeks gestational age at entry (p= >0.9999, unpaired t test). The difference in the mean gestational age at entry was statistically insignificant (p >0.05).

PRE-TREATMENT EFW (gms)



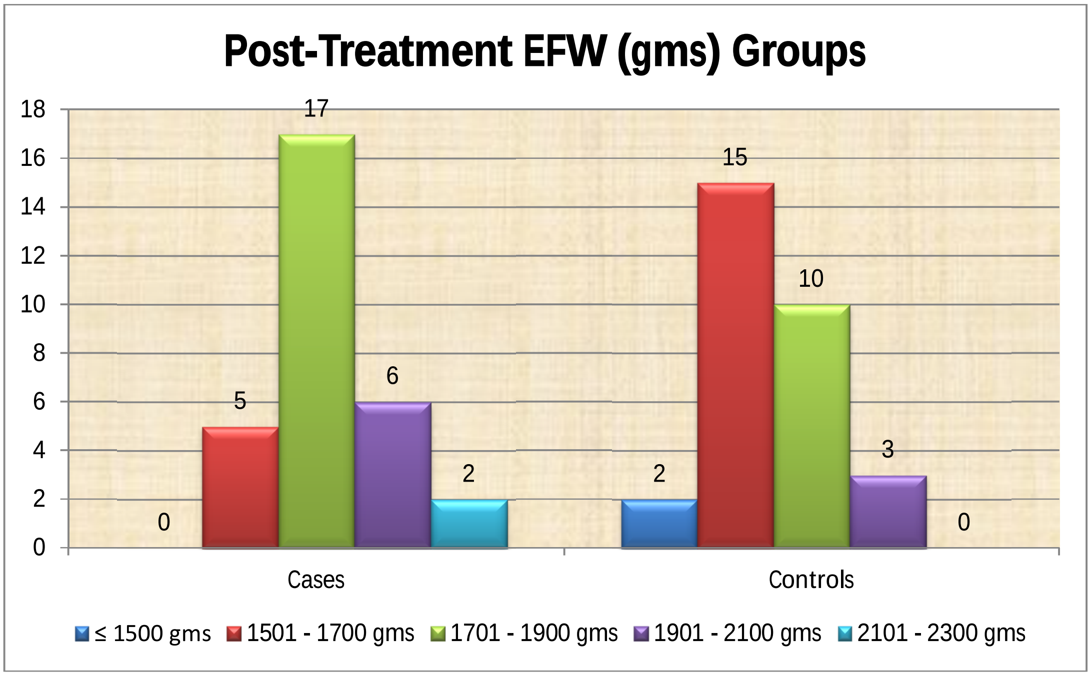
Pre-Treatment EFW (gms) Groups	Cases	%	Controls	%
≤ 1100 mgs	3	10.00	3	10.00
1101-1300 gms	16	53.33	14	46.67
1301-1500 gms	6	20.00	7	23.33
1501-1700 gms	5	16.67	6	20.00
Total	30	100.00	30	100.00



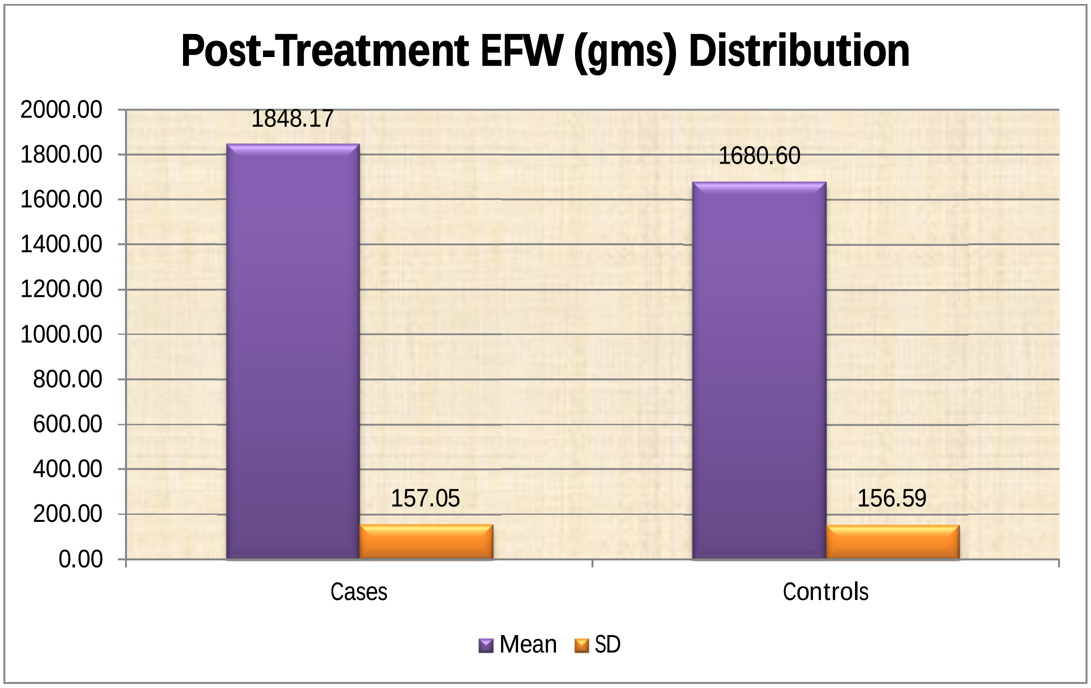
Pre-Treatment EFW (gms) Distribution	Cases	Controls
Mean	1288.97	1291.57
SD	159.23	166.50
P value (Unpaired t Test)		0.9509

The study subjects were distributed in 1101-1300 gms pre-treatment EFW group in cases group (n=10, 53.33%) and same pre-treatment EFW group in control group (n=14, 46.67%) (p=0.9509, unpaired t test), p value being insignificant.

POST-TREATMENT EFW (gms)



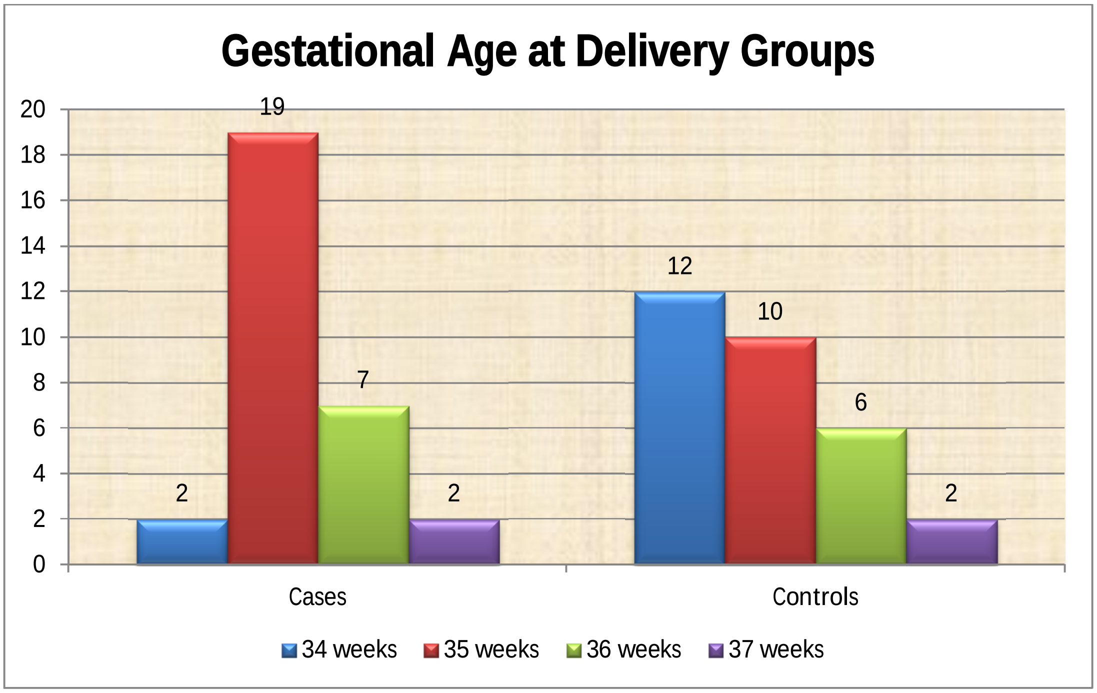
Post-Treatment EFW (gms) Groups	Cases	%	Controls	%
≤ 1500 gms	0	0.00	2	6.67
1501-1700 gms	5	16.67	15	50.00
1701-1900 gms	17	56.67	10	33.33
1901-2100 gms	6	20.00	3	10.00
2101-2300 gms	2	6.67	0	0.00
Total	30	100.00	30	100.00



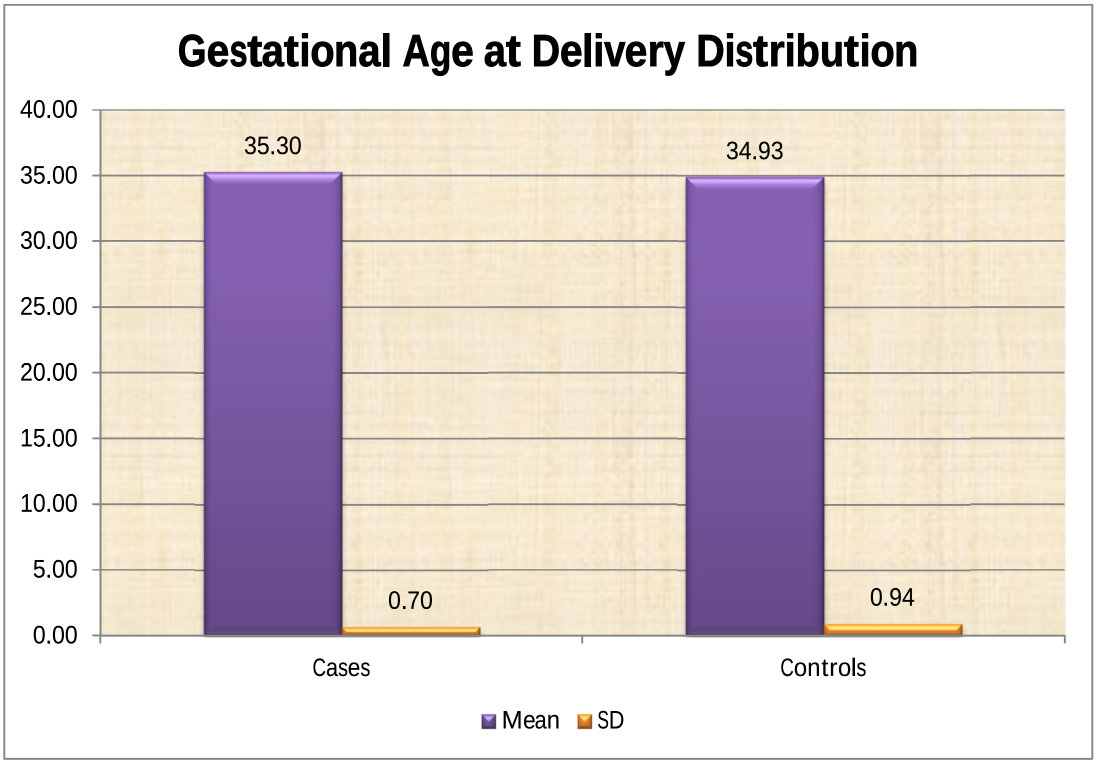
Post-Treatment EFW (gms) Distribution	Cases	Controls
Mean	1848.17	1680.60
SD	157.05	156.59
P value (Unpaired t Test)		0.0001

Cases group (n=17, 56.67%) 1701-1900 gms and in control group 1501-1700gms (n=15, 50.00%) (p=0.0001, unpaired t test). The difference in the mean post-treatment EFW in cases group (1848.17) and control group (1680.60) was found to be statistically significant ($p < 0.05$). The increased difference in the mean post-treatment EFW (167.57, 9% higher) was statistically significant ($p < 0.05$).

GESTATIONAL AGE AT DELIVERY



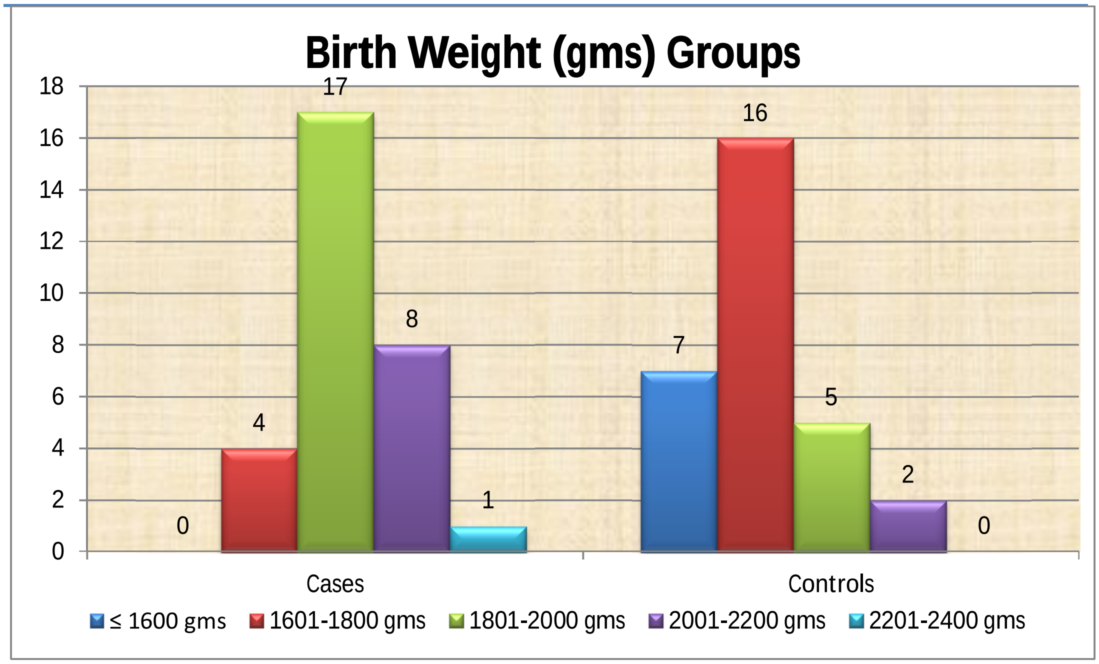
Gestational Age at Delivery Groups	Cases	%	Controls	%
34 weeks	2	6.67	12	40.00
35 weeks	19	63.33	10	33.33
36 weeks	7	23.33	6	20.00
37 weeks	2	6.67	2	6.67
Total	30	100.00	30	100.00



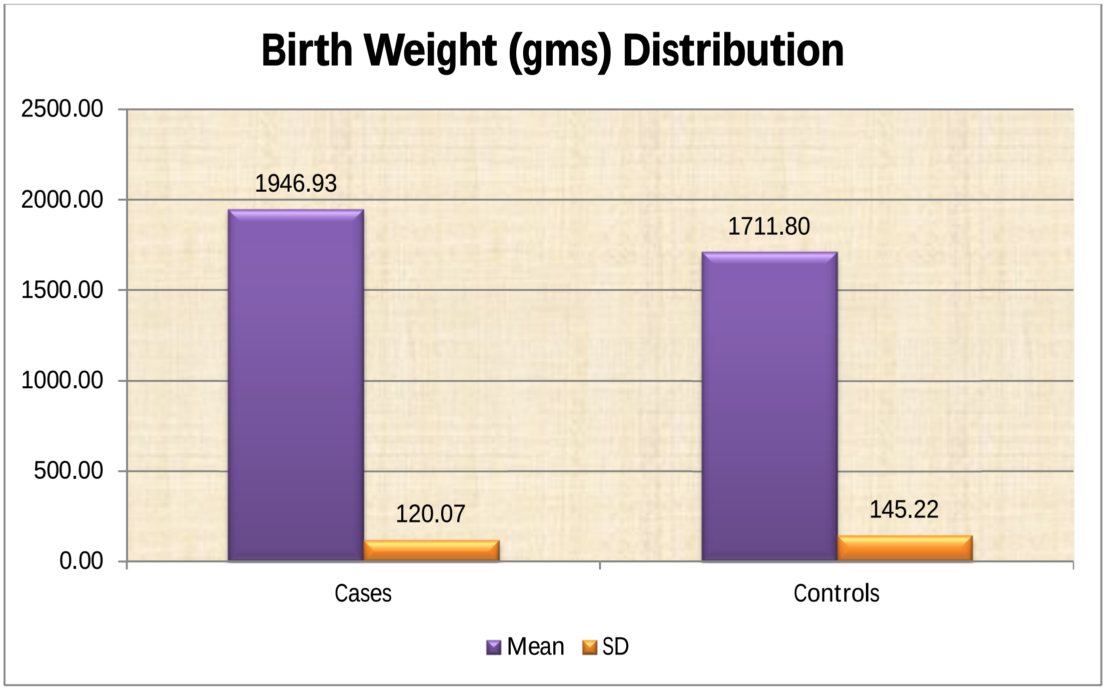
Gestational Age at Delivery Distribution	Cases	Controls
Mean	35.30	34.93
SD	0.70	0.94
P value (Unpaired t Test)		0.0933

Gestational age at delivery in case group was 35 weeks (n=19, 63.33%) and 34 weeks in control group (n=12, 40.00%) (p= 0.0933, unpaired t test), the difference being statistically insignificant (p >0.05).

BIRTH WEIGHT



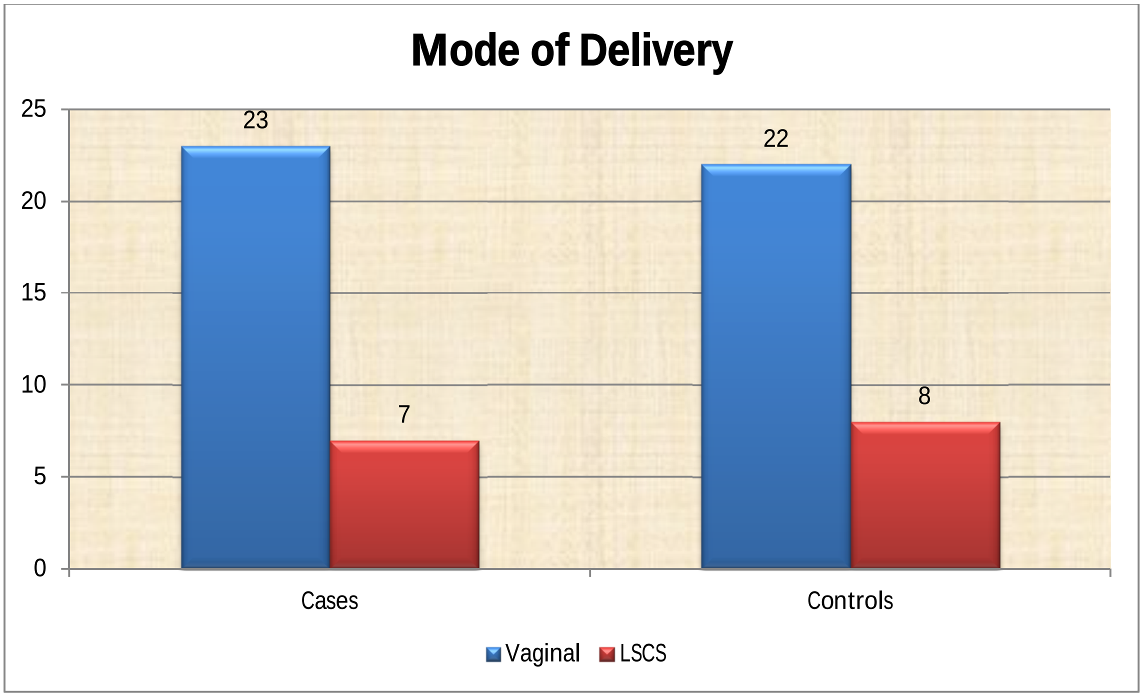
Birth Weight (gms) Groups	Cases	%	Controls	%
≤ 1600 gms	0	0.00	7	23.33
1601-1800 gms	4	13.33	16	53.33
1801-2000 gms	17	56.67	5	16.67
2001-2200 gms	8	26.67	2	6.67
2201-2400 gms	1	3.33	0	0.00
Total	30	100.00	30	100.00



Birth Weight (gms) Distribution	Cases	Controls
Mean	1946.93	1711.80
SD	120.07	145.22
P value (Unpaired t Test)		<0.0001

In cases group birth weight was in the range of 1801-2000gms group (n=17, 56.67%) and in control group 1600-1800gms (n=16, 53.33%) (p=0.0001, unpaired t test). The difference in the mean birth weight in cases group (1946.93) and control group (1711.80) was found to be statistically significant (p <0.05). The increased difference in mean birth weight (235.13gms, 12% higher) was found to be statistically significant (p <0.05).

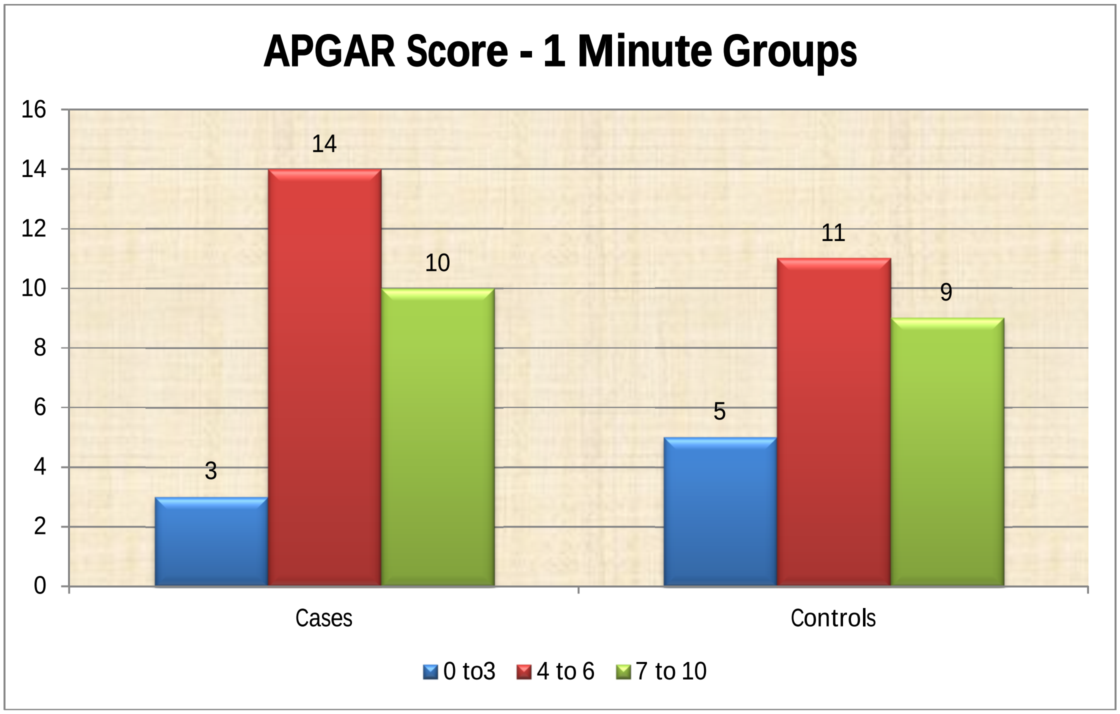
MODE OF DELIVERY



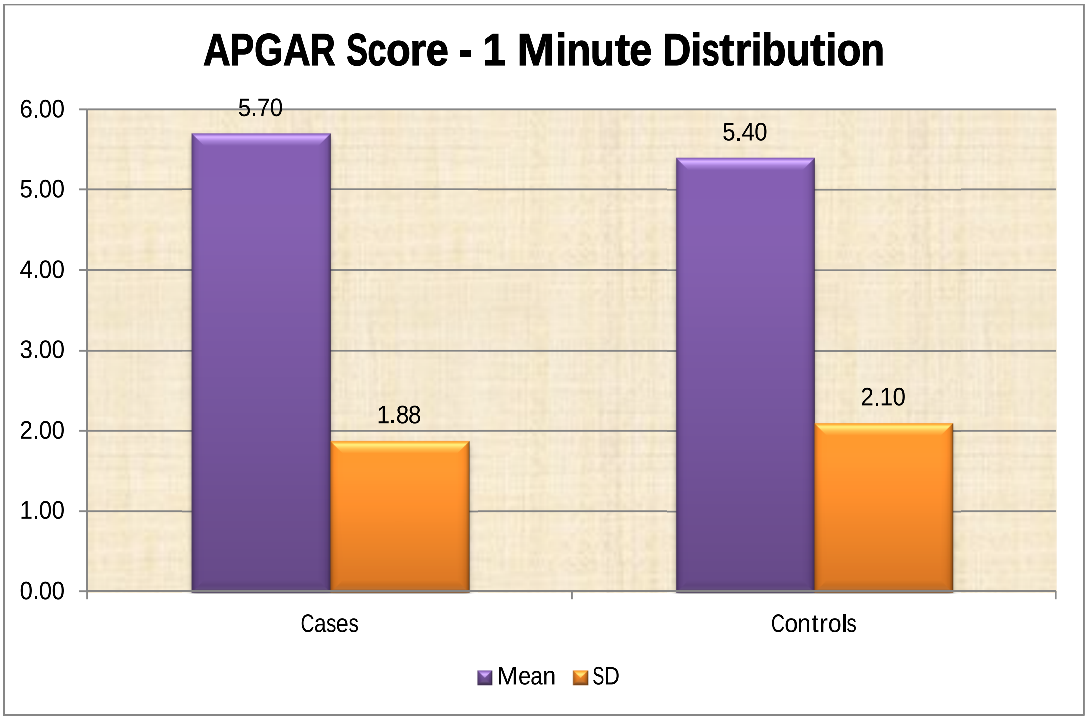
Mode of Delivery	**Cases**	**%**	**Controls**	**%**
Vaginal	23	76.67	22	73.33
LSCS	7	23.33	8	26.67
Total	30	100.00	30	100.00
P value (Chi Square Test)			0.7663	

The increased percentage difference in vaginal delivery rate in cases group and control group (3.33, 4 % higher) was found to be statistically insignificant (p >0.05).

APGAR SCORE - 1 MINUTE



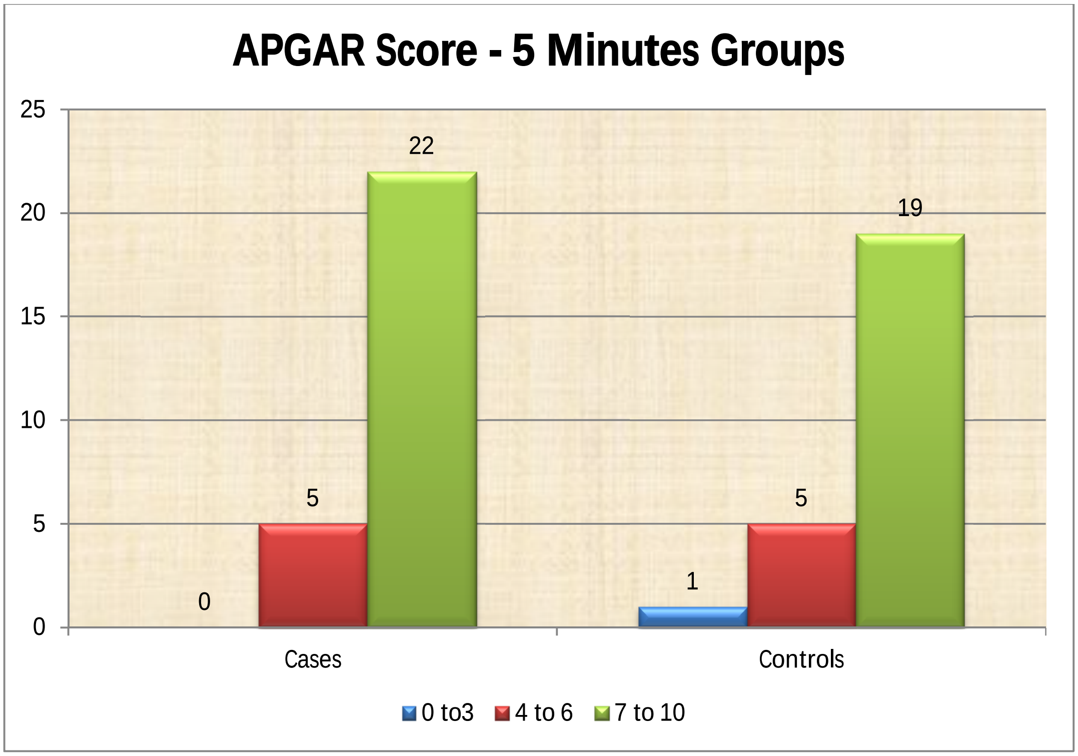
APGAR Score - 1 Minute Groups	Cases	%	Controls	%
0 to3	3	11.11	5	20.00
4 to 6	14	51.85	11	44.00
7 to 10	10	37.04	9	36.00
Total	27	100.00	25	100.00



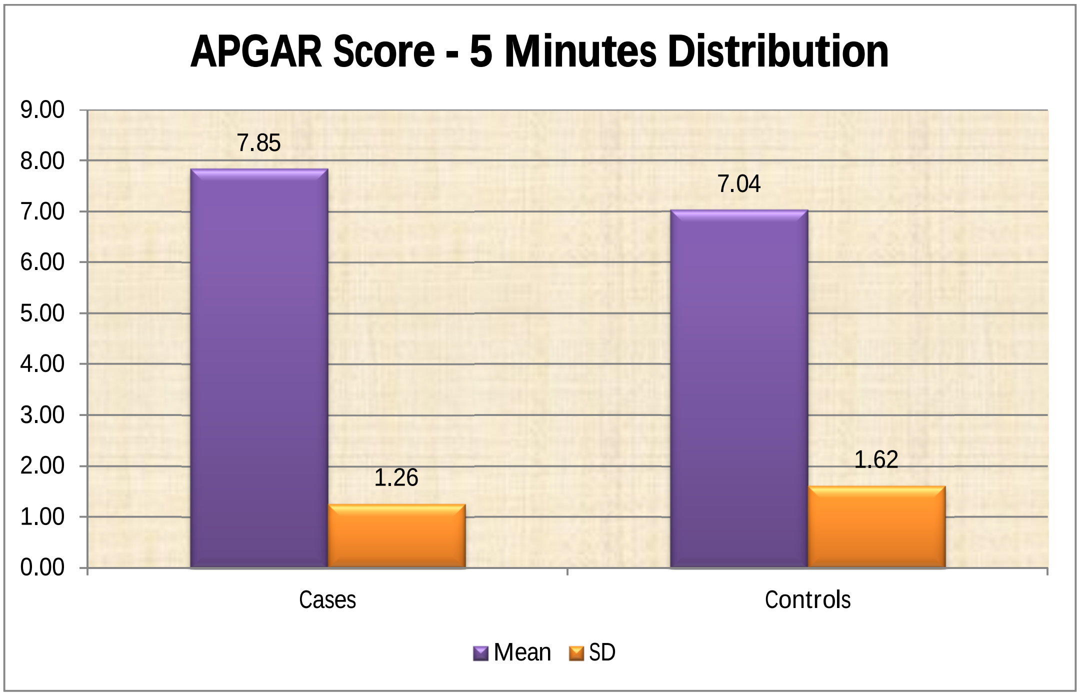
APGAR Score – 1 Minute Distribution	Cases	Controls
Mean	5.70	5.40
SD	1.88	2.10
P value (Unpaired t Test)		0.5845

The difference in the mean APGAR score at 1 minute in cases group (5.70) and control group (5.40) was found to be statistically insignificant ($p > 0.05$).

APGAR SCORE - 5 MINUTES



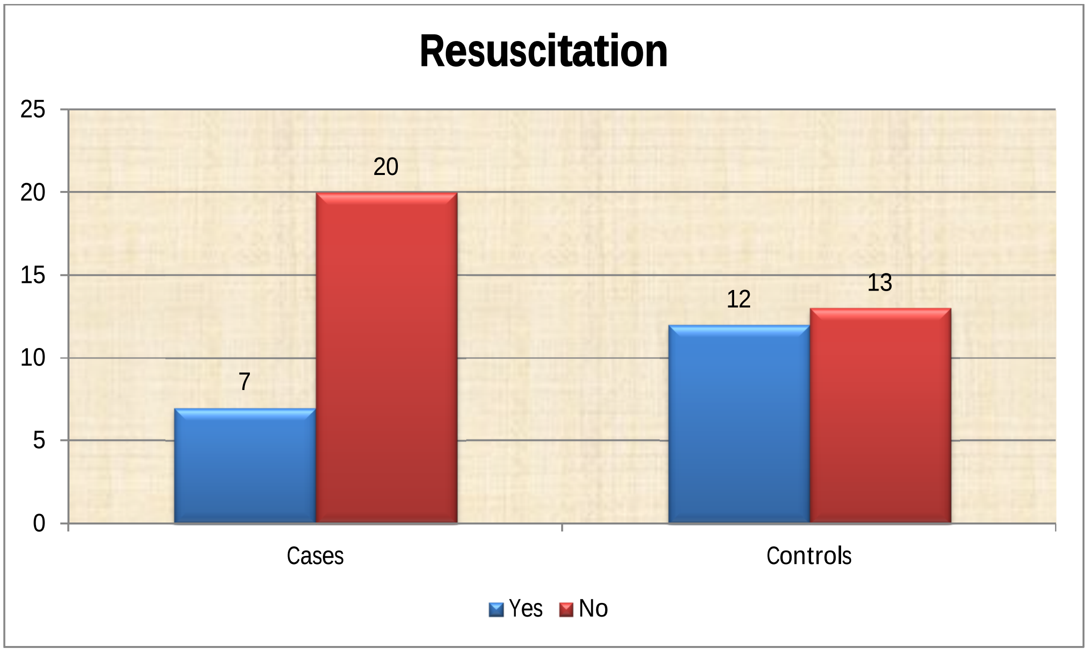
APGAR Score - 5 Minutes Groups	Cases	%	Controls	%
0 to3	0	0.00	1	4.00
4 to 6	5	18.52	5	20.00
7 to 10	22	81.48	19	76.00
Total	27	100.00	25	100.00



APGAR Score – 5 Minutes Distribution	Cases	Controls
Mean	7.85	7.04
SD	1.26	1.62
P value (Unpaired t Test)		0.0483

The range of 7-10 APGAR score at 5 minutes in case group (n=22, 81.48%) and same APGAR score at 5 minutes in control group (n=19, 76.00%) (p=0.0483, unpaired t test) The difference in the mean APGAR score at 5 minutes in cases group (7.85) and control group (7.04) was found to be statistically significant (p <0.05).

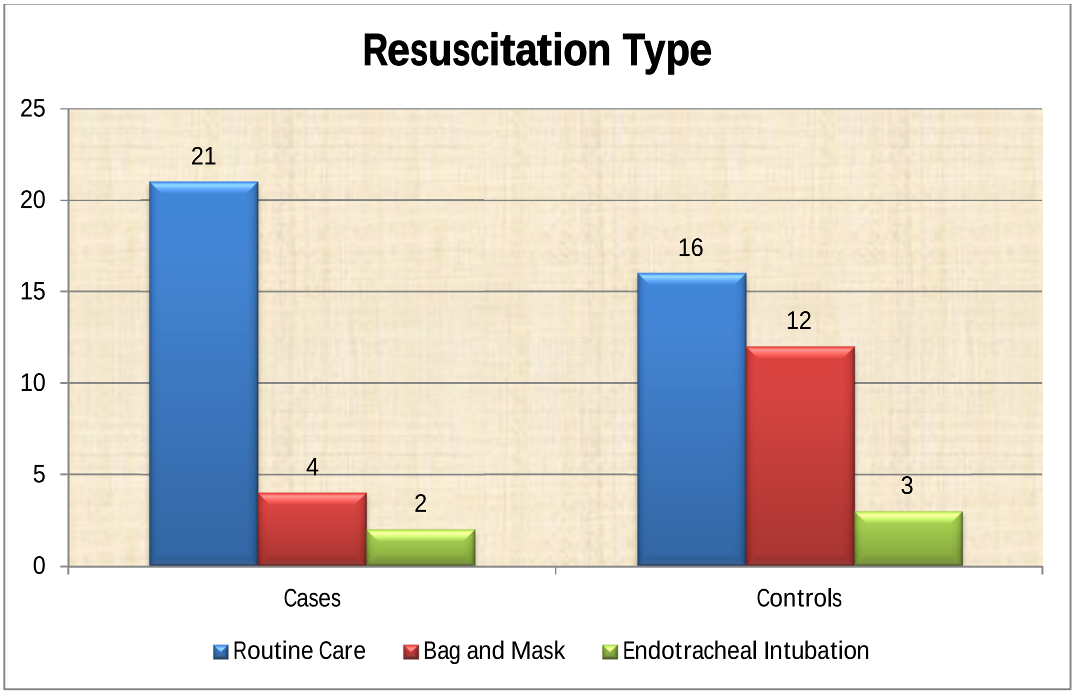
RESUSCITATION



Resuscitation	Cases	%	Controls	%
Yes	7	25.93	12	48.00
No	20	74.07	13	52.00
Total	27	100.00	25	100.00
P value (Chi Squared Test)			0.0991	

Resuscitation status of the study subjects is normally distributed across the intervention groups and the difference was statistically insignificant(p>0.05)

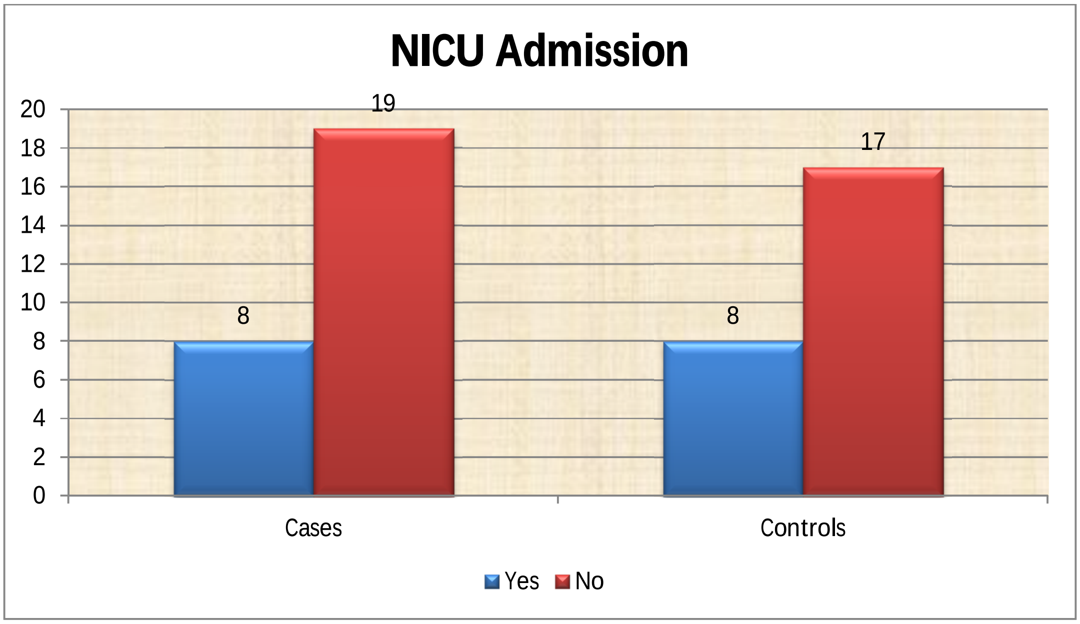
RESUSCITATION TYPE



Resuscitation Type	Cases	%	Controls	%	P value Chi Squared Test
Routine Care	21	77.78	16	64.00	0.2733
Bag and Mask	4	14.81	12	48.00	02114
Endotracheal Intubation	2	7.41	3	12.00	0.5752

The increased percentage difference in various resuscitation type in cases group and control group was found to be statistically insignificant (p >0.05).

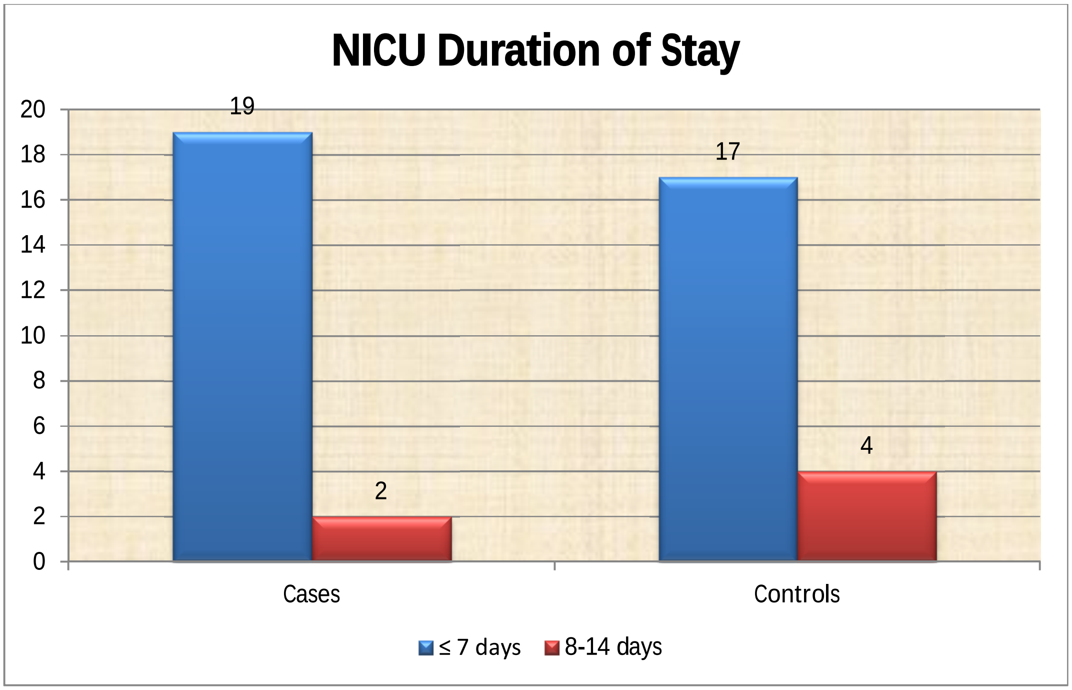
NICU ADMISSION



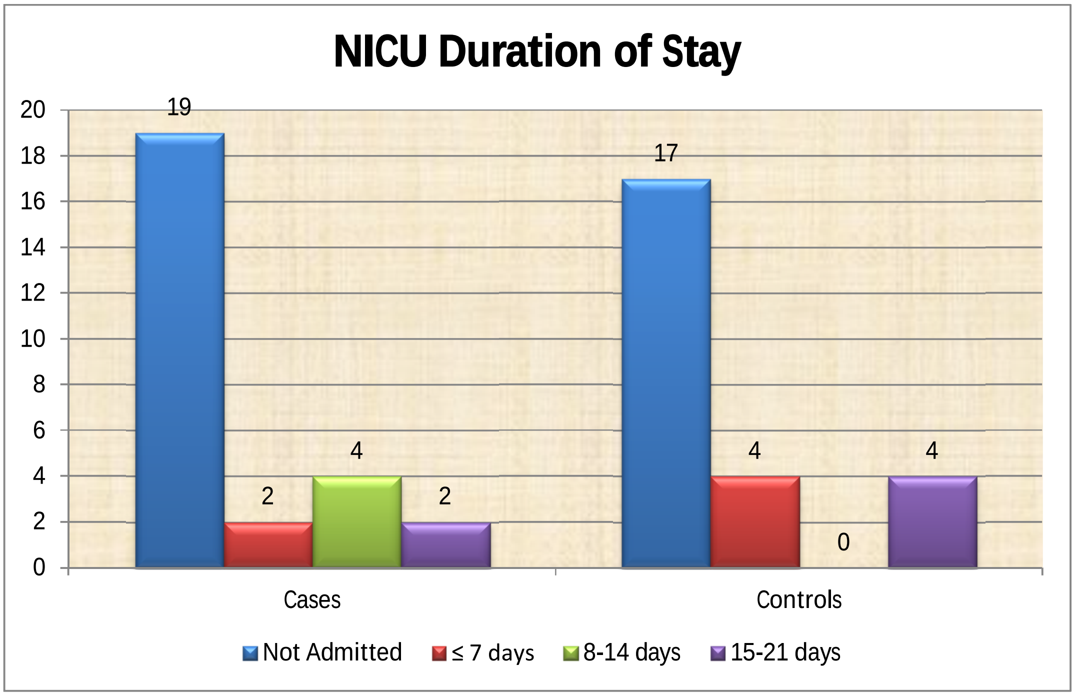
NICU Admission	Cases	%	Controls	%
Yes	8	29.63	8	32.00
No	19	70.37	17	68.00
Total	27	100.00	25	100.00
P value (Chi Squared Test)			0.9254	

The decreased percentage difference in NICU admission in cases group and control group (2.37, 7% lower) was found to be statistically insignificant (p >0.05).

NICU DURATION OF STAY



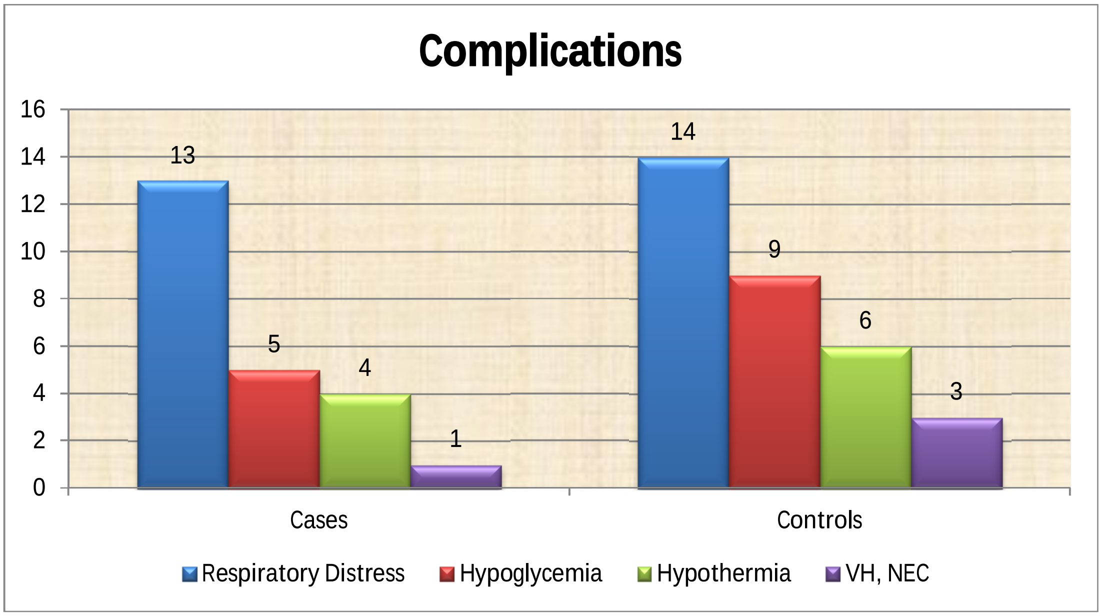
NICU Duration of Stay	Cases	%	Controls	%
Not Admitted	19	70.37	17	68.00
≤ 7 days	2	7.41	4	16.00
8-14 days	4	14.81	0	0.00
15-21 days	2	7.41	4	16.00
Total	27	100.00	25	100.00



NICU Duration of Stay Distribution	Cases	Controls
Mean	9.88	11.00
SD	4.19	7.05
P value (Unpaired t Test)		0.7039

The difference in the mean NICU duration of stay in cases group (9.88) and control group (11.00) was found to be statistically insignificant ($p > 0.05$).

COMPLICATIONS



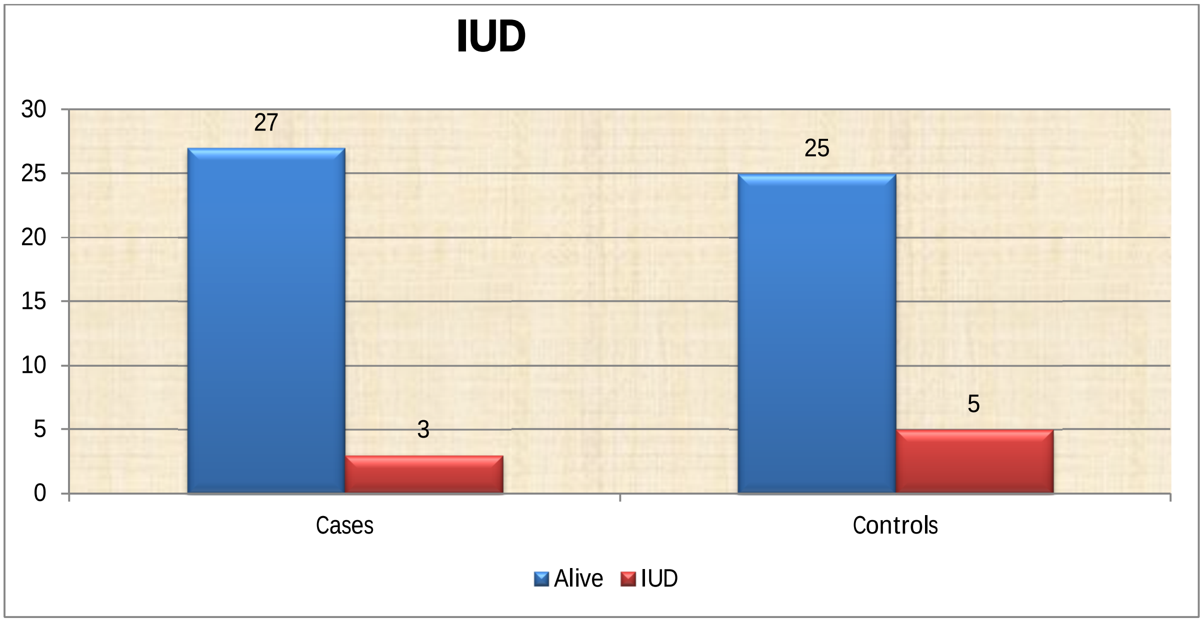
Complications	Cases	%	Controls	%	P value Chi Squared Test
RD	13	48.15	14	56.00	0.3712
Hypoglycemia	5	18.52	9	36.00	0.1567
Hypothermia	4	14.81	6	24.00	0.4011
VH, NEC	1	3.70	3	12.00	0.2622

RD – Respiratory distress   VH – Ventricular Hemorrhage

NEC – Necrotising enterocolitis

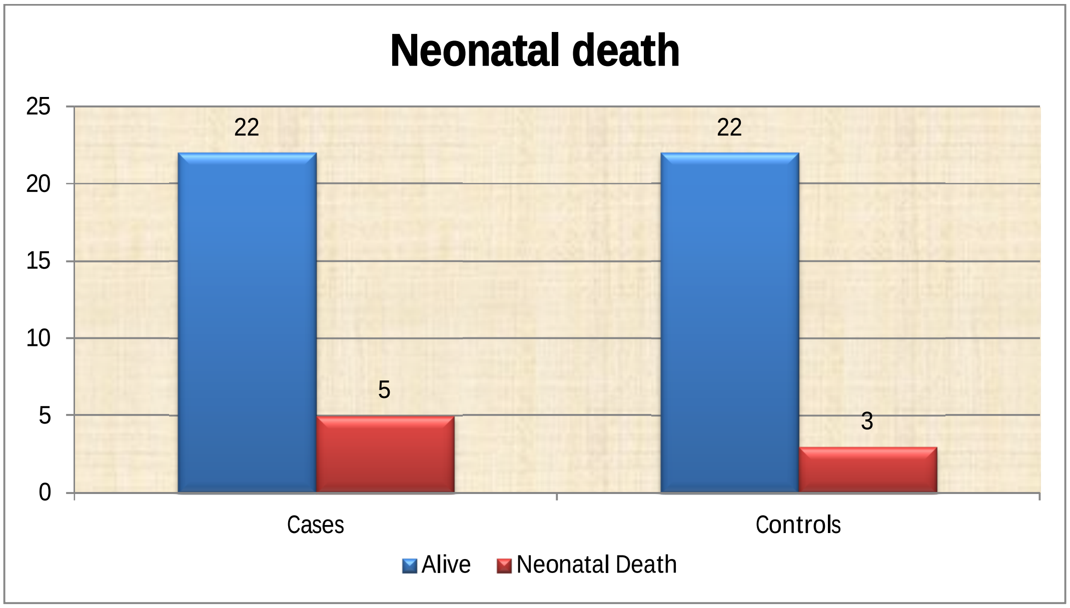
The decreased percentage difference in complication status in cases group and control group (10.70, 33% lower) was found to be statistically insignificant (p >0.05).

MORTALITY



Mortality rate	Cases	%	Controls	%
Live birth	27	90.00	25	83.33
IUD	3	10.00	5	16.67
Total	30	100.00	30	100.00
P value (Chi Squared Test)			0.4479	

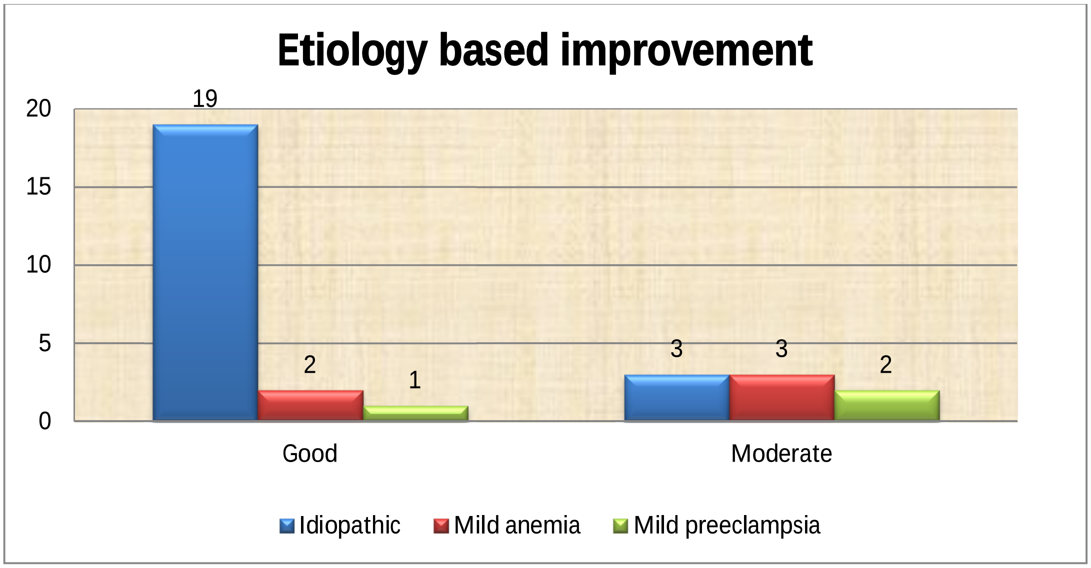
IUD rate in the cases group was 10% and in control group was 16.67% (p= 0.4479, chi squared test). The decreased percentage in IUD in case group was found to be statistically insignificant (p >0.05).



Mortality	Cases	%	Controls	%
Alive	22	81.48	22	88.00
Neonatal Death	5	18.52	3	12.00
Total	27	100.00	25	100.00
P value (Chi Squared Test)			0.5152	

Neonatal death in the cases group was 18.52% and in control group was 12%(p= 0.5152, chi squared test). The increased percentage difference in neonatal death status in cases group was found to be statistically insignificant (p >0.05).

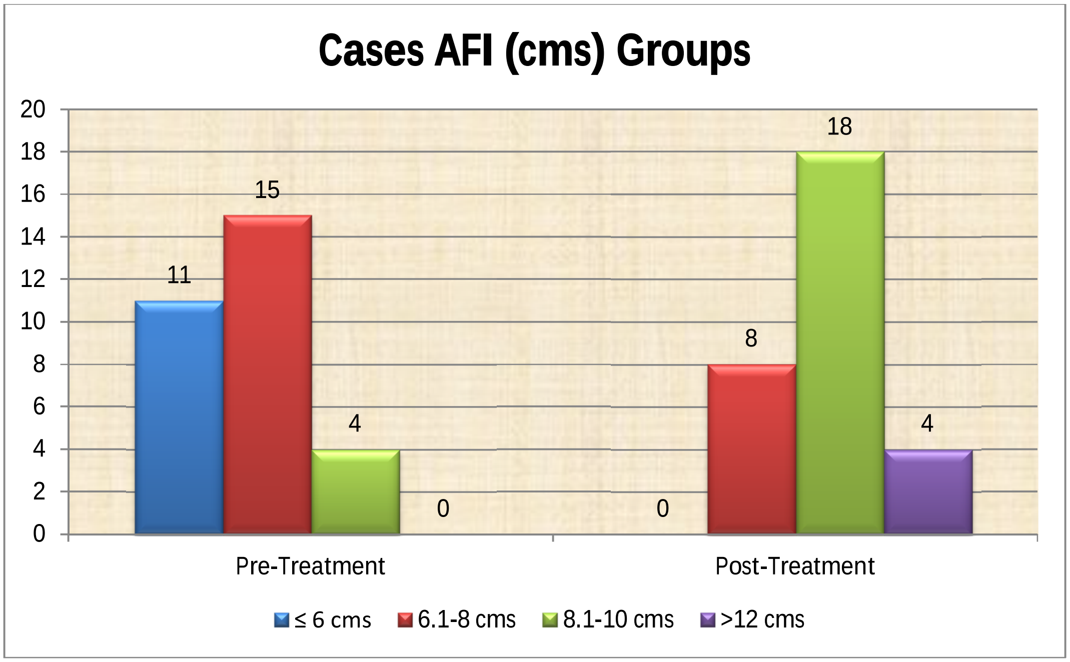
ETIOLOGY BASED IMPROVEMENT



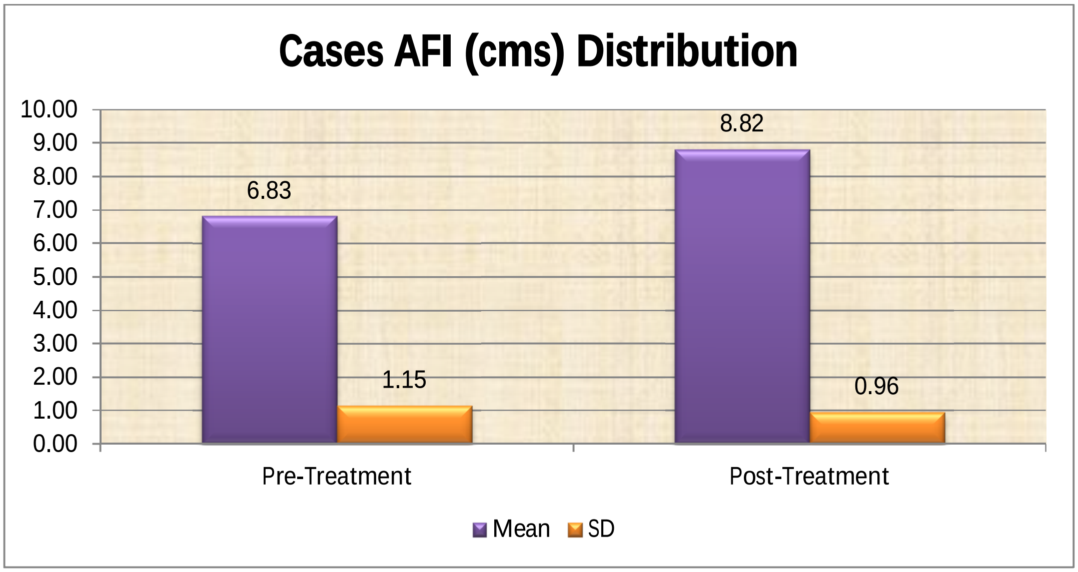
Cause	No. Of cases	Improvement of IUGR status	
		Good (100%)	Moderate(80%)
Idiopathic	22	19	3
Mild anemia	5	2	3
Mild preeclampsia	3	1	2
P value (Chi squared test)			0.02725

A significant p value in comparison of etiology based IUGR improvement indicates the significant improvement in idiopathic IUGR.

CASES AFI (cms)



Cases AFI (cms) Groups	Pre-Treatment	%	Post-Treatment	%
≤ 6 cms	11	36.67	0	0.00
6.1-8 cms	15	50.00	8	26.67
8.1-10 cms	4	13.33	18	60.00
>12 cms	0	0.00	4	13.33
Total	30	100.00	30	100.00



Cases AFI (cms) Distribution	Pre-Treatment	Post-Treatment
Mean	6.83	8.82
SD	1.15	0.96
P value (Paired t Test)		<0.0001

Pre intervention AFI 6.1-8 cms (n=15, 50.00%) and post intervention AFI 8.1-10 cms (n=18,60.00%) (p<0.0001, paired t test). The increased difference in mean post-treatment AFI (1.99, 23% higher) was found to be statistically significant (p <0.05).

Discussion

DISCUSSION

In my study, majority of the study subjects in cases group were distributed in 26-30 years age group (n=20,66.67%) and same age group in control group (n=15, 50.00%). The difference in the mean age of patients in cases group (27.77%) and control group (27.70%) was found to be statistically insignificant (p >0.05). In both the groups in the study, gestational age at entry was around 30 weeks, in case group (n=14, 46.67%) and in control group (n=14, 46.67%), with the difference in the mean gestational age at entry in cases group (30.60) and control group (30.60) being statistically insignificant (p >0.05).

CHARACTERISTICS	GROUP I	GROUP II	p value
Age	27± 3	27±2.7	>0.05
BMI	20±3	20±4	>0.05
Period of gestation	30±1	30±1	>0.05

Characteristics in study and control groups

Pre-treatment EFW was similar in both the groups between 1101-1300 gms in case group (n=10, 53.33%) and in control group (n=14, 46.67%). The post-treatment EFW analysed in both the groups, after a period of 4 weeks, showed 1701-1900 gms (n=17, 56.67%) in cases group and 1501-1700 gms in control group (n=15, 50.00%). The difference in the mean in cases group (1848.17) and control group (1680.60) (p <0.05) and the increased difference in mean post-treatment EFW in cases group compared to control group (167.57, 9% higher) was found to be statistically significant (p <0.05). Similar results were by presented Sieroszewski at al. The ultrasound estimation of fetal weight at the start and at the end of the treatment showed a mean increase of 642 g. By comparison, within the control group a mean value increase of 395 g (SE 77 g) was found. There was a significant statistical difference when comparing the estimated fetal weight increase.

CHARACTERISTICS	GROUP I		GROUP II		p value
	No	%	No	%	
Live births	27	90	25	83.33	>0.05
IUD	3	10	5	16.67	>0.05
Mean birth weight	30	1.9±0.12	30	1.7±0.14	<0.05
GA at delivery	30	35±0.7	30	34.9±0.94	>0.05
Vaginal delivery	23	76.67	22	73.33	>0.05
LSCS rate	7	23.33	8	26.67	>0.05

Outcome in study and control groups

Percentage of live birth was found to be more in Arginine therapy group (90 % in case group vs 83.33 % in control group). In case group, intrauterine deaths were 3 (10%) and there were 5 in control group (16.67%) ($p>0.05$), difference being statistically insignificant. Of the above, there were 5 neonatal deaths in case group (18.52%) and 3 in control group (12%), with a statistically insignificant p value.

Though the gestational age at delivery was found to be more in Arginine therapy group around 35weeks whereas it was around 34 weeks in control group, the difference was statistically insignificant.

The mean birth weight of the neonates in case group was 1801-2000 gms(56.67%) compared to 1601-1800gms(53.33%) in control group($p<0.05$). This implies that the difference in the mean birth weight in cases group (1946.93gms) and control group (1711.80gms) and the increased difference in mean birth weight of 235.13gms in cases group compared to control group (12% higher) was found to be statistically significant ($p <0.05$). This outcome was comparable to the study done by Xiao XM et al in 2005. Their study showed a significantly higher mean birth weight in group supplemented with Arginine ($P<0.05$) than in control group.

The incidence of vaginal delivery was 76.67% in cases group and 73.33% in control group. P value was found to be statistically insignificant ($p >0.05$).

Postnatal assessment showed that APGAR score at 1st and 5th minute was higher in the L-Arginine group. However, the difference between the APGAR scores at 1st minute in neonates of both the groups was statistically insignificant. APGAR score at 5 minutes in the range of 7-10 was distributed as follows: in case group(81.48%) and in control group (76.00%). On analysing the difference in the mean APGAR score at 5 minutes in cases group (7.85) and control group (7.04), it was found to be statistically significant ($p <0.05$). The increased difference in mean APGAR score at 5 minutes in cases group compared to control group (10% higher) was similar to the study done by Mariola Rapocka et al in 2007.

Neonatal outcome	Characteristics	Group I		Group II		P value
		No	%	No	%	
APGAR score	At 1 min					
	0 -3	3	11.11	5	20	>0.05
	4-6	14	51.85	11	44	>0.05
	7-10	10	37.04	9	36	>0.05
	At 5 min					
	0-3	0	0	1	4	<0.05
	4-6	5	18.52	5	20	<0.05
	7-10	22	81.48	19	76	<0.05
Complications	Respiratory Distress	13	48.15	14	56.00	>0.05
	Hypoglycemia	5	18.52	9	36.00	>0.05
	Hypothermia	4	14.81	6	24.00	>0.05
	VH, NEC	1	3.70	3	12.00	>0.05
Resuscitation	Routine	21	77.78	16	64	>0.05
	BMV	4	14.81	12	48	>0.05
	ET	2	7.41	3	12	>0.05
NICU admission		8	29.63	8	32	>0.05
NICU duration of stay		9.88±4.19		11 ±7.05		>0.05
Mortality		5	18.52	3	12.00	>0.05

Neonatal outcome in study and control group

Around 74.07% neonates in case group required no resuscitation measures at birth apart from routine care and it was 52.00% in control group but this difference was statistically insignificant($p>0.05$)

29.63% and 32.00% of the neonates of case group and control required NICU admission and had a hospital stay of 9.88 ± 4.19 and 11 ± 7.05 days respectively, difference being statistically insignificant. 48.15% of neonates in case group and 56.00 % in control group had respiratory distress after birth. The frequency of hypoglycaemia in cases group and control group were 18.52 and 36.00 % respectively. Hypothermia was seen in 14.81 % of neonates of case group and 24.00 % of neonates of control group. The incidence of ventricular haemorrhage and necrotizing enterocolitis was 3.70 % in neonates of case group compared to 12.00% in control group thus suggesting a lower incidence of these complications in Arginine therapy group, although this difference was insignificant statistically. The outcome was comparable to the study by shalini singh et al.

When etiology was considered and analysed, women with idiopathic IUGR, i.e. those having nutritional deficiency responded well to L-Arginine therapy than those with anemia or preeclampsia. Even on correction of mild preeclampsia, the difference in the weight improvement was insignificant.

Interesting results according to fetal growth were presented by Rytlewski's group in 2007. They analyzed a group of patients with preeclampsia. Preeclampsia increases the risk of intrauterine growth restriction, likely by impairment of fetomaternal blood flow. In their group patients, treatment with exogenous L-Arginine significantly increased newborn APGAR score and duration of pregnancy.

A secondary outcome that was measured in my study was the improvement of liquor status with L-Arginine in the patients. This variable was analysed only in the cases group where we supplemented L-Arginine. In control group, women with severe oligohydramnios were not included as they could not be given L-Arginine and it raised ethical issue. In pre-treatment status, the women in cases group were distributed in 6.1-8 cms AFI group (50.00%) and 8.1-10 cms AFI group (60.00%). The difference in the mean pre-treatment AFI group (6.83) and post-treatment AFI group (8.82) was found to be statistically significant ($p < 0.05$). The result was similar to the study done by Hebbar et al on L-Arginine effect on liquor status.

Conclusion

CONCLUSION

The conclusion drawn from my study is that after oral administration of L-Arginine, in women where fetal well being is good and no placental insufficiency as indicated by decreased Umblical Artery S/D ratio, there was increase in the birth weight of the baby, improved APGAR of the neonates and thereby good perinatal outcome. There was reduction in complications and need for NICU admissions. L-Arginine improves foetal weight more significantly in cases with idiopathic IUGR or where mother is nutritionally deficient rather than in those pregnancies affected by anemia or preeclampsia. Anemia should be corrected in the preconception stage to improve pregnancy outcome. Hence, during antenatal care all pregnant women and high risk cases should be screened to detect IUGR in earlier stages which will decrease perinatal mortality and morbidity. Apart from the routine fetal surveillance in IUGR, Umbilical Artery S/D ratio done by doppler ultrasonography helps in detecting increased resistance and monitoring of a compromised fetus. The IUGR cases should be supplemented oral L-Arginine, a nitric oxide donor, to reduce the resistance in feto-placental circulation.

Bibliography

BIBLIOGRAPHY

1. Ropacka M, Kowalska J, Blumska-Hepner K, Markwitz W, Bręborowicz GH. Effect of L-arginine on fetal outcome in IUGR fetuses. Arch Perinat Med 2007;13:30-4.

2. Shalini Singh, Archana Singh, Deepika Sharma, Abha Singh, M. K. Narula, Jayashree Bhattacharjee. Effect of L-Arginine on Nitric Oxide Levels in Intrauterine Growth Restriction and its Correlation with Fetal OutcomeInd J Clin Biochem (July-Sept 2015) 0(3):298–304 DOI 10.1007/s12291-014-0450-4.

3. Dera A, Ropacka M, Kowalska J, Markwitz W, Nycz P, Bręborowicz GH. The effect of L-arginine treatment on the neonatal outcome from pregnancies complicated by intrauterine growth restriction and gestational hypertension. Arch Perinat Med 2007;13:35-9.

4. Lampariello C, De Blasio A, Merenda A, Graziano E, Michalopoulou A, Bruno P. Use of L-arginine in intrauterine growth retardation (IUGR): Authors' experience. Minerva Ginecol 1997;49:577-81.

5. Staff AC, Berge L, Haugen G, Lorentzen B, Mikkelsen B, Henriksen T.Dietary supplementation with L-arginine or placeboin women with pre-eclam-psia. Acta Obstet Gynecol Scand 2004;83:103-7.

6. RCOG (1996) Nitric oxide, the endothelium, pregnancy and preeclampsia.Br. J. Obstet. Gynaecol. 103: 4-15.

7. Weiner C.P., Baschat A.A., (1999) Fetal growth restriction: evaluationand management. James D.K., Steer P.J., Weiner C.P.,Gonik B., (Ed.) High-Risk Pregnancy: Management options, 2nd ed. Saunders, London, UK, 291-307.

8. Neri I., Mazza V., Galassi M.C. et al. (1996) Effects of L-arginineon utero-placental circulation in growth retarded fetuses.Acta Obstet. Gynecol. Scand. 75: 208-212.

9. Sieroszewski, P., Suzin J., Karowicz-Bilinska A. (2004) Ultrasoundevaluation of intrauterine growth restriction therapy bya nitric oxide donor (L-arginine). J. Matern. Fetal Neonatal Med.5: 363-366.

10. Facchinetti F., Longo M., Piccini F., Neri I. (1997) Arginine load in preeclampsia: a test of nitric oxide reserve. Prenat. Neonat.Med. 2: 7-10.

11. Rytlewski K., Olszanecki R., Lauterbach R. Et al. (2006) Effectsof Oral L-Arginine on the Foetal Conditio and Neonatal outcomein Preeclampsia: a preliminary Report. Pharmacol. Toxicol. 99:146-152.

12. Di Iorio R., Marinoni E., Caocci F. Et al. (1997) Amniotic fluidnitric oxide and uteroplacental blood flow in pregnancy complicatedby intrauterine growth retardation. Br. J. Obstet. Gynecol.104: 1134-1139.

13. Xiao X.M., Li L.P. (2005) L-arginine treatment for asymmetric fetal growth restriction. J. Gynecol. Obstet. 88: 15-18.

14. Wu G, Morris SM Jr. Arginine metabolism: nitric oxide and beyond. Biochem J. 1998;336:1–17.

15. Wu G, Bazer FW, Wallace JM, Spencer TE. Board-invited review intrauterine growth retardation: implications for the animal sciences. J Anim Sci. 2006;84:2316–37.

16. Grisham MB, Johnson GG, Lancaster JR Jr. Quantitation of nitrite and nitrate in extra-cellular fluids. Methods Enzymol. 1996;268:237–46

17. Thureen PJ, Baron KA, Fennessey PV, Hay WW Jr. Ovine placental and fetal arginine and metabolism at normal and increased maternal plasma arginine concentrations. Peads Res. 2002;51:464–71.

18. Montagnani M, Chen H, Barr VA, Quon MJ. Insulin-stimulated activation of eNOS is independent of Ca2? but requires phosphorylation by Akt at Ser1179. J Biol Chem. 2001;276:30392–8.

19. Lakhkar BN, Rajagopal KN, Gaurishankar PT. Doppler prediction of adverse perinatal outcome in PIH and IUGR. Indian J Radiol Imag. 2006;16(1):109–16.

20. Roy A, Mukherjee S, Bhattacharyya SK, Banerjee P, Das B, Patra KK. Perinatal outcome in pregnancies with intra-uterine growth restriction by using umbilical and middle cerebral artery colour Doppler. J Indian Med Assoc. 2012;110(3):154–7.

Annexure

PROFORMA

Name- Phone No-
Age-
OP No-
Address-
Socioeconomic status-

MENSTRUAL HISTORY: Regular / irregular, LMP

MARITAL HISTORY: Married since ___ years

OBSTETRIC HISTORY : Parity-
History of abortion-
No of living children-
Last child birth-

PRESENTING COMPLAINTS (IFANY)-
PAST HISTORY- Diabetes
Hypertension
Heart disease
Tuberculosis
Epilepsy
Chronic renal failure
Obesity
Drug intake
Previous history of IUGR
FAMILY HISTORY –
TREATMENT HISTORY-
ALLERGY HISTORY-

<u>EXAMINATION:</u>
Height-
Weight- current : pre-pregnancy :
BMI- current : pre-pregnancy :
Temp-
Pulse rate -
Blood pressure-
Pallor-

Pedal edema-
Symphysiofundal height -

CVS-
RS-
Per Abdomen-

BLOOD INVESTIGATIONS:
Blood grouping :
HIV- VDRL- HBSAG-
OGCT –
COMPLETE BLOOD COUNT:
Hb
TC
DC
ESR
Platelets
RFT:
Blood Sugar-
Blood Urea-
S.Creatinine-
URINE
Albumin-
Sugar-
LFT:
SB-
TP-
Alb-
Glo-
SGOT-
SGPT-
ALP-
ULTRASONOGRAPHY:
Dating Scan :
Anomaly Scan :
Recent Scan :
(Liquor status
& GA with doppler)

URKUND

Urkund Analysis Result

Analysed Document: THESIS FINAL (Autosaved).docx (D31021125)
Submitted: 10/4/2017 8:00:00 PM
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https://www.omicsonline.org/open-access/postnatal-complications-of-intrauterine-growth-restriction-2167-0897-1000232.pdf
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CERTIFICATE – II

This is to certify that this dissertation work titled **"L-ARGININE SUPPLEMENTATION IN IUGR AND IT'S EFFECT ON FETAL OUTCOME"** of the candidate **Dr. Hajee Arshiya Bareen** with registration Number **221516153** for the award of **M.S., Degree** in the branch of **OBSTETRICS AND GYNAECOLOGY**. I personally verified the urkund.com website for the purpose of plagiarism Check. I found that the uploaded thesis file contains from introduction to conclusion pages and result shows **13%** percentage of plagiarism in the dissertation.

Guide & Supervisor sign with Seal.

INSTITUTIONAL ETHICS COMMITTEE
GOVT. KILPAUK MEDICAL COLLEGE,
CHENNAI-10
Protocol ID. No.06/2016 Meeting held on 14/12/2016
CERTIFICATE OF APPROVAL

The Institutional Ethical Committee of Govt. Kilpauk Medical College, Chennai reviewed and discussed the application for approval **"L-ARGININE SUPPLEMENTATION IN IUGR AND ITS EFFECT ON FETAL OUTCOME"** submitted by Dr.Hajee.Arshiya Bareen.,Post Graduate in MS O&G ,Govt. Kilpauk Medical College, Chennai.

The Proposal is APPROVED.

The Institutional Ethical Committee expects to be informed about the progress of the study any Adverse Drug Reaction Occurring in the Course of the study any change in the protocol and patient information /informed consent and asks to be provided a copy of the final report.

6/1/17

DEAN
Govt. Kilpauk Medical College,
Chennai-10.

4/1/17

MASTER CHART -CASES (L-ARGININE SUPPLEMENTED)

S.NO	NAME	AGE	IP.NO	GESTATIONAL AGE AT ENTRY	BODY MASS INDEX (BMI)	PRE-TREATMENT EFW (in kg)	PRETREATMENT AFI (in cms)	POST TREATMENT EFW(AFTER 4 WEEKS) (in kg)	POST TREATMENT AFI (in cms)	GESTATIONAL AGE AT DELIVERY	BIRTH WEIGHT(in Kg)	MODE OF DELIVERY (VAGINAL-V / LSCS-L)	APGAR SCORE		RESUSCITATION(YES-Y /NO-N)	RESUSCITATION TYPE (YES -Y / NO-N)			NICU ADMISSIONS	DURATION OF STAY (NA - NOT APPLICABLE)	COMPLICATIONS (YES-Y/NO-N)				NEONATAL MORTALITY (YES-Y/NO-N)
													AT 1 MIN	AT 5 MIN		ROUTINE CARE	BAG AND MASK	ENDOTRACHEAL INTUBATION			RESPIRATORY DISTRESS	HYPOGLYCEMIA	HYPOTHERMIA	VH, NEC	
1	Kokila	31	35229	30	24.1	1190	6	1710	8.1	35w5d	1915	V	8	9	N	Y	N	N	N	NA	N	N	N	N	N
2	Manimala	26	30426	33	22.8	1689	8	2238	10.1	37w4d	2254	L	3	8	Y	N	Y	N	Y	9	Y	Y	N	N	Y
3	Deepa	28	30356	30	23.7	1185	6.2	1756	8	36w	1978	V	6	9	N	Y	N	N	N	NA	N	N	N	N	N
4	Anuradha	25	32972	32	21.9	1518	8	2068	10	36w5d	2079	V	4	7	Y	Y	N	N	N	NA	N	N	N	N	N
5	Jeevalakshmi	30	32982	30	22.5	1186	8.5	1699	10.3	35w5d	1927	V	2	6	Y	N	N	Y	Y	15	Y	N	Y	N	Y
6	Sandhiya	26	31358	30	17.8	1179	6.6	1711	8.2	34w5d	1720	V	5	9	N	Y	N	N	N	NA	N	N	N	N	N
7	Kalaivani	28	32897	31	25.8	1319	6	1876	8.15	35w6d	1890	V	8	9	N	Y	N	N	N	NA	Y	N	N	N	N
8	Pandi selvi	26	32671	30	27.1	1188	9	1764	10.5	34w6d	1800	L	7	8	N	Y	N	N	N	NA	N	N	N	N	N
9	Nishanthini	24	32789	31	24.5	1328	6.9	1899	9.1	35w4d	1906	V	1	5	Y	N	N	Y	Y	15	Y	Y	Y	Y	Y
10	Devi	29	32145	30	26.7	1197	6.2	1751	8.4	35w1d	1755	V	5	8	N	Y	N	N	N	NA	Y	N	N	N	N
11	Sivagami	31	32190	30	22.9	1210	8.2	1845	10	35w	1996	L	8	9	N	Y	N	N	N	NA	N	N	N	N	N
12	Vidhya	28	29344	29	21.6	1098	5.5	1677	7.8	35w2d	2072	V	6	9	N	Y	N	N	N	NA	Y	N	N	N	N
13	Jeyasri	30	29814	32	23.7	1524	6	2016	8.3	36w2d	2020	V	6	9	N	Y	N	N	N	NA	N	N	N	N	N
14	Gayathri	27	30410	31	22.4	1299	8	1872	9.7	35w	1880	V	5	7	N	Y	N	N	N	NA	Y	N	N	N	N
15	Eswari	25	29103	30	24	1198	5.8	1723	8.1	35w6d	1965	L	8	9	N	Y	N	N	N	NA	N	N	N	N	N
16	Palani	26	30575	30	24.4	1210	9	1790	10.4	35w	1978	V	7	8	N	Y	N	N	N	NA	N	N	N	N	N
17	Sasikala	21	30658	31	21.7	1343	7	1913	9	35w5d	1920	V	5	6	Y	N	Y	N	Y	9	Y	Y	Y	N	Y
18	Seetha	28	30662	32	22.3	1511	6.2	2070	8.1	36w1d	2075	V	4	6	Y	N	Y	N	Y	8	Y	N	Y	N	Y
19	Nithyakalyani	29	30625	30	18.1	1199	7.9	1652	9.9	35w1d	1761	V	5	7	N	Y	N	N	Y	13	Y	N	N	N	N
20	Manju	39	30280	31	29.7	1311	5.75	1867	8.05	35w4d	1870	V	7	8	N	Y	N	N	N	NA	N	N	N	N	N
21	Priya	31	30619	29	20.7	1090	7.2	1611	9	35w	1932	V	5	7	N	Y	N	N	N	NA	Y	N	N	N	N
22	Dhanalakshmi	27	30662	30	23.2	1195	8	1879	9.8	35w6d	2014	L	6	9	N	Y	N	N	N	NA	N	N	N	N	N
23	Kotteeswari	30	30635	31	29.6	1299	6.1	1785	8	36w	1889	V	8	9	N	Y	N	N	N	NA	N	N	N	N	N
24	Shoba	29	30453	29	20.8	1087	5.7	1649	7.9	35w3d	1931	V	5	6	Y	N	Y	N	Y	7	Y	Y	N	N	N
25	Sangeetha	26	30634	30	22	1222	8	1836	9.6	35w5d	1890	L	8	9	N	Y	N	N	N	NA	N	N	N	N	N
26	Thilagavathi	24	30631	30	23.1	1212	7	1798	9	35w	1831	L	7	8	N	Y	N	N	N	NA	N	N	N	N	N
27	Kalpana	29	30614	32	24	1432	5.6	2010	7.9	36w6d	2014	V	5	7	N	Y	N	N	Y	3	Y	Y	N	N	N
28	Sulochana	26	30008	33	22.1	1690	5.4	2199	7.7	37w4d	2200	V	IUD												
29	Saranya	29	267798	30	21	1236	5.82	1871	7.92	36w6d	2034	V	IUD												
30	Jeyachitra	25	29845	31	28.9	1324	5.3	1910	7.6	35w1d	1912	V	IUD												

MASTER CHART -CONTROLS (CONVENTIONAL TREATMENT)

S.NO	NAME	AGE	IP.NO	GESTATIONAL AGE AT ENTRY	BODY MASS INDEX (BMI)	PRE-TREATMENT EFW (in kg)	POST TREATMENT EFW(AFTER 4 WEEKS) (in kg)	GESTATIONAL AGE AT DELIVERY	BIRTH WEIGHT(in Kg)	MODE OF DELIVERY (VAGINAL-V / LSCS-L)	APGAR SCORE		RESUSCITATION (YES-Y /NO-N)	RESUSCITATION TYPE (YES -Y / NO-N)			NICU ADMISSIONS	DURATION OF STAY (NA-NOT APPLICABLE)	COMPLICATIONS				NEONATAL MORTALITY (YES-Y/NO-N)
											AT 1 MIN	AT 5 MIN		ROUTINE CARE	BAG AND MASK	ENDOTRACHEAL INTUBATION			RESPIRATORY DISTRESS	HYPOGLYCEMIA	HYPOTHERMIA	VH, NEC	
1	kaviya	25	33182	30	21.2	1194	1580	34w3d	1604	L	3	5	Y	Y	Y	N	Y	18	Y	Y	Y	N	N
2	amrutha	27	29171	29	22	1091	1486	34w2d	1512	V	6	7	N	Y	N	N	N	NA	N	N	N	N	N
3	semmozhi	32	31101	33	24.1	1687	2063	37w5d	2070	V	1	4	Y	N	Y	Y	Y	5	Y	Y	Y	Y	Y
4	shivani	26	31101	30	17.8	1189	1585	35w1d	1620	L	8	9	N	Y	N	N	N	NA	N	N	N	N	N
5	ayesha	31	32351	31	23.8	1321	1725	35w	1725	V	5	7	Y	N	Y	N	Y	17	Y	Y	N	N	N
6	nivetha	25	33168	30	21.5	1198	1603	35w3d	1675	V	7	8	N	Y	N	N	N	NA	N	N	N	N	N
7	boomika	24	30917	31	29	1328	1730	35w1d	1731	V	5	7	Y	N	Y	N	N	NA	Y	N	N	N	N
8	eelavarasi	31	33176	32	19.9	1519	1892	36w1d	1895	V	4	6	Y	N	Y	N	Y	18	Y	Y	N	N	N
9	kasthuri	30	30188	29	23.2	1094	1487	34w1d	1523	L	6	8	N	Y	N	N	N	NA	N	N	N	N	N
10	akalya	29	33281	30	20	1194	1601	34w2d	1610	V	8	9	N	Y	N	N	N	NA	Y	N	N	N	N
11	smithika	26	32159	30	28.9	1191	1597	34w	1597	V	7	8	N	Y	N	N	N	NA	N	N	N	N	N
12	anandhi	24	33314	31	21.4	1319	1715	35w2d	1723	V	2	4	Y	N	Y	Y	Y	6	Y	Y	Y	Y	Y
13	suganya	29	31139	29	24	1095	1503	34w1d	1601	L	5	7	Y	N	Y	N	N	NA	Y	N	N	N	N
14	mallarvilli	32	30217	30	30.1	1192	1579	34w2d	1584	V	6	8	N	Y	N	N	N	NA	N	N	N	N	N
15	vaasini	31	30281	32	18.1	1531	1886	36w	1898	V	7	8	N	Y	N	N	N	NA	N	Y	N	N	N
16	nadia	29	31662	31	22.6	1340	1713	35w2d	1718	V	8	9	N	Y	N	N	N	NA	N	N	N	N	N
17	geethika	25	33417	31	22	1337	1739	35w	1740	L	5	7	Y	Y	Y	N	N	NA	Y	N	N	N	N
18	chandrika	29	32146	30	19.1	1191	1596	34w6d	1620	V	1	3	Y	N	Y	Y	Y	2	Y	Y	Y	Y	Y
19	vidya	24	33681	30	27	1178	1584	35w2d	1652	V	7	8	N	Y	N	N	N	NA	N	N	N	N	N
20	devika	26	30317	31	22.6	1338	1705	35w2d	1710	L	6	7	N	Y	N	N	N	NA	N	N	N	N	N
21	dhanyaa	28	29289	30	21	1193	1573	34w6d	1598	V	5	7	Y	Y	Y	N	N	NA	Y	N	N	N	N
22	harika	27	30699	30	23.7	1192	1565	36w1d	1742	V	3	6	Y	N	Y	N	Y	5	Y	Y	Y	N	N
23	shabareen	31	31845	32	21	1527	1903	36w1d	1908	L	8	9	N	Y	N	N	N	NA	N	N	N	N	N
24	praveena	29	33711	30	24.1	1194	1589	35w2d	1692	V	7	8	N	Y	N	N	N	NA	Y	N	N	N	N
25	rehana	24	32965	33	25.9	1690	2060	37w3d	2062	L	5	7	Y	N	Y	N	Y	17	Y	Y	Y	N	N
26	shwetha	26	33567	30	23	1192	1592	34w3d	1599	V	IUD												
27	kamatchi	32	33617	32	31	1524	1888	36w	1892	V	IUD												
28	bharathi	24	30507	30	21	1188	1589	34w5d	1596	V	IUD												
29	venuka	28	31106	31	19.7	1330	1702	36w	1823	V	IUD												
30	chandana	27	32787	30	28.9	1190	1588	34w6d	1634	V	IUD												